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Over the course of the last century, a successful history of fire suppression has contributed to unsuccessful present day control over wildfire. In the absence of fire and the janitorial and ecological services it provides, drier inland forests are shifting in species composition and exceeding densities that cannot survive and persist in current fire patterns occurring on the landscape. In the past two decades, managers have introduced restoration use of fire and thinning for their ecological benefits and to convert fuel-heavy forests to fuel-lean landscapes to lessen the threat of stand-replacing wildfire. In this study, we evaluated the long-term impact of forest restoration practices on soil biochemistry and the mycorrhizal fungi associated with ponderosa pine (*Pinus ponderosa)*. Study sites were located in the Blue Mountains of northeastern Oregon where treatments implemented in 1998 and 2000 included mechanical thinning of forested areas, prescribed fire, a combination of thinning followed by fire, and an untreated control. Soil sampling for this study occurred in 2014 and included four replications of each treatment for a total of 16 experimental units. Fungal-specific sequence analysis of morphotyped root-tips from ponderosa pine was used to assess the species diversity and root-tip density abundance of each fungal taxon within each sample unit. Additionally, litter depth, pH, and soil carbon (C), nitrogen (N), and Bray phosphorus (P) were measured from soil cores that were stratified into depths of 0-5cm and 5-10cm. Bray-P, pH, and percent C differed among treatments. Bray-P and percent C and N differed between soil depths. The results indicated that given a decade-plus period of recovery, mycorrhizal fungi in dry inland forests dominated by ponderosa pine returned to levels similar to the untreated controls. Similar litter depths across treatments suggest that litter depth stabilizes over time in these forests. Soil C and nutrient differences may have been driven by the thinning treatments and the resultant deposition of residual slash following harvesting or the consumption of slash by prescribed fire. Elevated ectomycorrhiza biomass in the thinning and burning treatment may be a response of the host trees that creates a larger nutrient acquisition network in a less fertile environment. The results of this study demonstrate the resiliency of these forests to disturbances associated with restoration treatments, providing managers increased flexibility of options if maintaining abundant and persistent fungal communities is a concern. Given that the mean fire return interval for these forests is 15 years, a second reintroduction of prescribed fire may be timely.

Fuel Treatments of Ponderosa Pine (*Pinus ponderosa*) in the Blue Mountains of Eastern Oregon: A Mycorrhiza Perspective

by
Benjamin T. N. Hart

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I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

Benjamin T. N. Hart, Author

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CHAPTER 1: A HISTORY OF THE BLUE MOUNTAINS, FACTORS INFLUENCING THE CURRENT DILEMMA ON FEDERAL LANDS AND THE ROLE OF FUNGI IN RESTORATION

1. A History of the Blue Mountains and the Evolution of Fire Suppression

The Blue Mountains of Oregon encompass a geographic region over 4,000 square miles in size, spanning from Prineville, Oregon and then sweeping east and north into lower south-east Washington and a sub-range extending into Idaho. The Blue Mountains are a collective of geologic features called terranes including the Baker, Wallowa, Olds Ferry, Grindstone, and Izee and create in an inland island-arc formation (Vallier and Brooks 1995). Following a series of geologic uplifts, subductions, and deep excision by hydraulic processes, the Blue Mountains have a reputation for steep, arduous terrain with a wide spectrum of soil types and plant communities. The rain shadow created by the Cascade Mountain Range to the west limits the amount of precipitation that can move east across Oregon. Within the Blue Mountains, maritime influences are able to travel along the Columbia River gorge that facilitate forests in the northern portion of the ecoregion, while rangelands composed of Juniper and sage-steppe are more common to the south.

The forested regions of the Blue Mountains were historically composed of aggregate stands of Douglas-fir series forest-types on northern slopes where soils are deeper and ponderosa pine on southern slopes where soils are thinner. The primary source of disturbance on these landscapes were low-intensity fires that occurred in the late summer and fall at intervals of 20 years or less (Heyerdahl et al. 2001). Lightning ignitions were common along this “Lightning Ally” but during the pre-European era, human caused ignitions from indigenous peoples were also a regular occurrence to improve forage for game species in following years. Forest structure was typically composed of large diameter trees in scattered clumps in mosaics common in old-growth stands (Youngblood et al. 2004). These forests promoted development of large, old trees as the regular fire return interval maintained an open understory of smaller diameter trees that established between fires; either cambial girdling or crown scorch from low-intensity ground fire often removed smaller trees and shrubby understory plants. This early form of silviculture resulted in large, park-like stands of trees with an open understory and a primarily herbaceous forest floor dominated by fescue bunchgrass and shrubs such as snowberry, gooseberry and huckleberry (Langston 1995).

Indigenous peoples of the Blue Mountains included the Walla Walla, Cayuse and Umatilla Tribes. Permanent village sites were located in the river valleys and lower elevation valleys, and upland regions were utilized for subsistence living during the summer and early fall. As part of seasonal departures from upland living, it was common practice to ignite grasslands to burn uninterrupted into the fall when season ending precipitation occurred. This practice removed dead plant material and stimulated grasses to grow with increased vigor the following spring. Additional anthropogenic reasons for introducing fire was increasing production of plants for foods like camas, medicinal plants and herbs, fiber plants used in basketry, and forage species for game management (Pyne 1982; Anderson 2005).

The arrival of Europeans began in the early 19th century with the arrival of French fur trappers to the region, the beginning of the long series of anthropogenic changes to occur on the West's landscape in the coming century. The arrival of settlers began in the mid 19th century with reports from fur trappers of a fertile land rich with plenty of room for settlement. Settlers brought with them livestock and domesticated animals, and quickly began converting the land around them to make room for crop and livestock production.

While the impact of European settlement had a large impact on conversion of plant and forb communities due to grazing of cattle and sheep, the impact of large-scale commercial timber had a landscape effect on changing the structure of the forest. Forests that contained thousands of acres of large diameter ponderosa pine were clear-cut in the forested mountain west in the late 19th and early 20th century. According to Pyne (1982), slash generated from the commercial harvesting was one of the contributors to the devastating wildfires that occurred in the early 1900's. These fires, and particularly the massive and deadly fire of 1910 (the "Big Burn") burned over 2 million acres of forest in Idaho, Washington and part of Montana in less than 36 hours, claiming dozens of lives and millions of dollars in lost timber revenue (Donovan and Brown 2007). The public outcry for keeping communities safe and private timber influences on the federal government prompted the creation of the United States Forest Service whose primary objective was the management of forests for continued wood production and the suppression of all fires that threatened a national resource (Egan 2009). This fire suppression effort resulted in the creation of the "10am Policy" in which stated "that a fire was to be contained and controlled by 10 a.m. following the report of a fire, for, failing that goal, control by 10 a.m. the next day and so on" (Pyne 2004). During the early days of forest management and fire suppression, fires were generally of such low intensity that a single ranger on horseback with a shovel was all that was needed to contain a lightning ignition. Later, as automobiles became more prevalent and an expanding workforce emerged from the New Deal of 1933, it was made possible to suppress increasingly large fires. Suppression efforts enhanced following the end of WWII, as not only equipment for the war effort like planes, bulldozers, and radio equipment were becoming available as military surplus, but there was also a large able-bodied workforce of men who had been trained to work hard and follow orders. A militarized culture of fire suppression quickly developed, which has continued to influence response to fire on public lands and directed the tactics used to address it.

Coinciding with the shift in mentality to full suppression of all forest fires, there was the introduction of a public-relations icon that has also proved difficult to adapt to current understanding. Smokey Bear was originally conceptualized in 1945 and by 1947 the slogan "Remember, only you can prevent forest fires!" came into the public sphere. The effectiveness of the campaign to promote fire suppression efforts proved successful to a fault. As fire managers now strive to re-image the role of fire on the landscape, the public still has the ingrained image of fire seemingly "burned" into their minds.

However, the effects of the fire suppression campaign altered forest structure, and more importantly, wildfire behavior. Organic material, once periodically removed by fire, now accumulated on the forest floor. Additionally, shade-tolerant trees normally down-regulated by the presence of fire were now able to establish and create dense stands. These trees not only accelerated the process of organic material deposition, but also created a combustible connection from the ground to the canopy. Un-combusted material on the forest floor creates fuel beds that can smolder for long periods, damaging tree roots and causing mortality of belowground microorganisms. The increased moisture retention of the soil created by increased fuel beds altered tree rooting structure, encouraging growth towards the surface, where they would subsequently be scorched when fire did pass through (Langston 1995). Fires that historically were smaller and easier to contain were now growing larger and by the 1980's, costs of fire suppression were skyrocketing. Severe fires in 1980's and 90's stimulated dialogue among forest managers about potential proactive measures. Early pioneers for the re-introduction of fire included Dr. Harold Biswell working in ponderosa stands of northern California in the early 1950’s (Van Wagtendonk 1995) and Drs. Hienselman and Wright working in conifer stands during the 1970’s (Hienselman and Wright 1973). Their early stance on the beneficial role of fire initially encountered skepticism from managers who were not only risk-averse to prescribed fire escaping containment lines, but also held the viewpoint of forests being a commodity, and the academic concept of forests being part of a larger and highly complex ecosystem had yet to evolve. By the 1990’s, however, managers were desperate to produce methods to reduce negative effects of fire, while also producing sustainable timber volumes and promote fire-resilient stands of trees, i.e. trees that were larger with structural complexity and diversity that could serve a purpose beyond just production of wood products. Prescribed burning is common in the southeast and in parts of the Midwest to maintain grasslands and improve forage for game species, and the use of prescribed fire under specific weather conditions has become a primary method of restoring fire-suppressed landscapes by combusting organic matter from the forest floor and removing smaller diameter shade tolerant saplings and poles. The use of mechanical thinning is another method often employed to modify forested areas into a more fire-safe condition while producing wood products to meet financial targets of federal lands.

2. Fuels Reduction Treatments and Impact on the Landscape

Currently over 98% of fires are completely suppressed before they reach 120 hectares, but fires that exceed that size account for 98% of all suppression costs (Calkin et al. 2005). In an effort to take a more proactive stance on the role of fire on the landscape, federal land managers began seeking other methods of fire management, rather than waiting for fire to occur on the landscape and then addressing it. Thinning forested areas to reduce stand densities is more common, but prescribed burning also offers unique advantages.

Thinning forests is most routinely achieved with the use of mechanical harvesting equipment. This not only increases the efficiency of harvesting activities but also increases worker safety. Public acceptance of mechanical thinning is also generally higher because smoke related issues associated with prescribed fire are not a concern. One advantage to use of mechanized thinning is that there is more precise control over the structure of the forest once thinning treatments are completed. Trees can be retained based on their species, canopy characteristics, and arrangement within the stands. Operations are less seasonally restricted than prescribed burning, which can only occur at certain times of the year according to specific windows of temperature, humidity, wind and fuel-bed characteristics. If trees are large enough to be commercially thinned, then there is an increased incentive for an economic return on fuels reduction activities, making it appealing to commercial logging and subsequent revenue being generated by the forest selling timber contracts. However, if there are not trees large enough to be sold either commercially or to specialty wood product markets, the cost of thinning alone can exceed $2,500/hectare (Haines et al. 2001). Donovan and Brown (2007) estimate the cost of treating only the most critically needed forests as of 2005 to be over $12 billion, while the operating budget for the Forest Service and Department of Interior combined was around $450 million. Complete treatment would take over 20 years, and many of these forests have vegetation recovery that would require re-treatment within that window of time. While mechanical treatment accomplishes reducing stand density, it does not reproduce many of the ecological benefits of fire such as mobilization of nutrients from combusted fuels and creation of snags and patchy forest composition. Authors have also expressed concerns about issues around soil compaction (Jennings et al. 2012).

Prescribed burning is the intentional burning of forested and range areas under specific weather conditions with the intention of removing woody debris and other organic matter from the forest floor to mimic patterns that would occur normally during low-intensity fire events (Knapp et al. 2009). In the humid-temperate regions of the west, typical prescribed burning is conducted at times of the year when fire behavior will be the most moderate, either before or after the summer drought period. In the Blue Mountains, this usually means that burning is conducted after winter dormancy and snowmelt but before fuels have become too dry or after fuels have moistened with the first rains and temperatures/humidity have reached desirable levels.

Prescribed fire has benefits unique to it compared to mechanical thinning. Due to combustion of organic material and subsequent translocation of nutrients mobilized, there is an ephemeral pulse of nutrients into the soil such as occurs with natural low-intensity wildfire, maintaining ecological processes. Additionally, heat-girdling of the cambium of small diameter trees induces mortality and results in snag creation within the stand providing services to wildlife such as mammals and birds, and trees that later fall to the ground provide habitat for arthropods and other ground-dwelling organisms. The cost of prescribed fire is low compared to thinning, with average costs per hectare around \$200, while reported costs from the Interagency Fire Center in 2016 estimate the cost of wildfire suppression around \$715 (https://www.nifc.gov/). However, there is no means to recoup those monies once spent as in thinning efforts. Additionally, there is limited control in stand structure and tree species composition as there is in thinning. Lastly, the use of fire means that there is the risk of fire spreading to the canopy and burning trees desired for retention, or worse for fire to escape the containment line and turn into a wildfire. Regardless, specific burn conditions must be adhered to in order to minimize risk and ensure air quality standards do not create a nuisance to the public.

3. The Role of Collaboratives in Forest Restoration

Historically, decisions on management practices on Federal lands were made in a top-down fashion, minimizing public input from the process and not making needs specific to localized area a priority. Additionally, the historic role of federal land management agencies like the Forest Service was to make forest products available for commercial utilization, and not to manage the land for objectives valued today like conservation of species diversity and preserving opportunities for public use like recreation. This commodity based approach created distrust with members of the public concerned with conservation and protection of the environment. With the passage of the Northwest Forest Plan of 1994, the Federal agencies now had a mandate to preserve as much sustainable harvesting of trees as possible while also ensuring the long-term survival of the spotted owl within its known range (Tuchmann et al. 1996). This mandate included avoiding harvest of late seral trees to preserve habitat that the spotted owl and associated plant and wildlife communities depend on. This dramatically altered the old ways of timber harvesting and effectively stopped logging of old-growth trees, having major economic impact on members of the timber community and eroding trust in Federal land management practices. At the same time, members of the environmental protection community had been struggling to preserve these same forests. The lack of accelerated or more sweeping action by the Forest Service came across as a lack of sincere effort to respond effectively, degrading trust on this side of the issue as well. Management of Forest Service lands became increasingly difficult, where “declining budgets, complex regulations and long-standing multiple-use conflicts have led to gridlock and litigation” (Urgenson et al. 2016). The lack of communication has led to a culture of adversarial outlook to those on opposing sides of the issue.

Recently, there has been incentive to establish multi-party communication and cooperation in order to accelerate forest restoration projects, especially in the dry conifer forests prone to risks of wildfires. Beginning in 1992 on the Deschutes National Forest, a group of community members formed to address issues surrounding the management of these public lands. They were the model of what has grown to be a statewide and now regionally active movement called *Forest Collaboratives*. Collaboratives are defined as “an organized collection of landowners, stakeholders, resource agencies, tribes, or other organizations that come together to address common issues and resolve problems through deliberation, consensus-building, and cooperative learning” (Goldstein and Butler 2010).

The objective is to build trust and relationships between members of the collaborative so that consensus can be reached about what projects should be pursued on Federal land, agree on risks and rewards and what measures will be taken in order to ensure their success. Based on the success of collaborative efforts in places like the Blue Mountains, the Forest Service has steadily began to take on collaboratives and provide support around issues like fire management and forest restoration (Wondolleck and Yaffee 2003).

One of the foundational values of collaboratives is that “no one gets everything they want, but everyone gets something they want” out of the actions taken. By establishing action-plans, the chance of litigation and barriers during the federal project planning process diminish dramatically, and the trend to put blame on the Agency if a management strategy fails to meet its objective is reduced. The role of the collaborative is to identify reasonable project ideas, provide details about the project scope and scale, secure agreement to work together between stakeholders, provide alternative action plans, and remind members of their commitments and previous decisions.

As forest restoration continues to rise in popularity and the need to reduce fire severity over increasingly large landscapes continues to accelerate, there will be an accompanying need to decrease the amount of pushback from the public and increase cooperation between conflicting parties. One of the biggest strengths of collaboratives is that they create a sense of inclusiveness and facilitate the process of building trust between parties that historically held value-based disagreements. By building relationships and fostering understanding, the chance that forests will continue to be able to accommodate the myriad demands placed on them by society will increase. Projects that obtain goals such as fire resiliency and forest restoration will ideally result in work for members of the timber community, preservation of habitat, preserving intact forests for recreationalists, and sustaining clean water for communities and betterment of fish populations. The role of collaboratives will continue to be a crucial component of the process as they incorporate and in some cases even fund research on the science most relevant to their local areas so that management decisions are the most appropriate to the communities wants and the environments needs.

4. Mycorrhizae and their Role in Resilient Forests

Traditionally, fungi were thought to function exclusively in the role of decomposition of organic material and as parasites/pathogens to other organisms. It is true that decomposer fungi are largely responsible for the degradation of organic matter (particularly large diameter wood and trees), but the span of ecological function for fungi goes beyond just saprotrophy. Fungi also create non-lethal parasitic relationships, mutualisms including mutually beneficial symbiotic associations with plants. In the latter, fungi expand the functional rooting network of the host plant with which they associate and increase the ability of the host plant to absorb necessary attributes of the soil profile such as water and nutrients. This is known as *mycorrhizal symbiosis*. In exchange for the access to the increased root network by the small diameter hyphae of the fungi, the plant allocates carbon derived from its photosynthetic processes (Smith and Read 2010). The nearly universal symbiosis among land plants initiates from the time the seedling sprouts, and is crucial to the plants survival.

Two main classifications of mycorrhizal symbiosis are arbuscular(endo-) mycorrhizas and ectomycorrhizas. Arbuscular mycorrhizas (AM) are the primary type among seed plants, associated with over 97% of the species of plants found across the globe (Myer 1973). They are classified by the presence of arbuscules, which are located within the root cell cortex, along with storage vesicles found within and between cells, and are diagnostic for AM symbiosis (Smith and Read 2010). Ectomycorrhizas (ECM) are much less prevalent in terms of the number of species of plants with which they associate, but they predominately associate with woody perennials (such as members of the Families Pinaceae and Fagaceae), contributing to their vast geographic distribution and dominance of sheer land mass occupied. ECM are characterized by the presence of three structural components, the first being fungal mycelia that begin to envelop the growing tree root, and eventually completely isolate the root from the soil environment, creating what is known as the *mantle*. Secondly, mycelium also penetrate the outer root cortex via intercellular growth, where the development of the *Hartig net* occurs. This interface between the plant cells and the mycelial cells allows for transmission of the nutrients each symbiotic partner has to offer. The last being the vast network of fungal hyphae that make nutrient acquisition possible, the *extraradical* mycelium. The role of mycorrhizal fungi in the cycling of nutrients from organic to inorganic forms, readily assimilated by plants, directly influences the fertility and structure of soil and is essential to maintaining soil health and long-term productivity (Smith and Read 2010

The importance of this symbiosis cannot be overstated, but there is an additional caveat to this unique relationship that merits consideration. In the early 1980's, researchers began to think of the added benefit of having previously colonized plants in close proximity to newly planted individuals in order to accelerate the process of symbiotic initiation (Perry et al. 1982). This linkage was discussed by other authors (Molina and Trappe 1982, Molina et al. 1992, Miller and Allen 1992, Newman 1988), but it was not until the mid 1990's that it was actually demonstrated that shared mycorrhizal connections between trees resulted in exchange of nutrients via the mycelial connection (Simard et al. 1997a; Simard et al. 1997b). These connections, or *common mycorrhizal networks*, are a novel example of fungi acting as brokers between trees, whether it was between trees of different species or between overstory trees and their seedlings. In either case, growth and survival increased as a result of the connections, and were diminished without them.

The role of overstory trees assisting the survival of seedlings is important in any forested system, but can have particular use in forests experiencing regular disturbance, especially from fire. Shade intolerant trees that can support seedlings following a low severity burn will benefit from restoration activities. Often, the purpose of restoration treatments in fire-adapted systems is to maintain an intact forest, but to reduce competitive stress associated with overly dense stands. Historically, ponderosa pine stands were composed of large trees in low densities, which maintained an environment of little to no competition for the above ground canopy, and particularly the belowground rooting network (Bright 1914). Given that the canopy can be used as an effective approximation for the trees root network size (Smith 1964), subsequent increases in canopy growth due to release from light competition would result in increased root growth. This increased root growth would not only benefit the overstory tree by creating larger reserves of belowground carbon, but would benefit future seedling recruitment by expanding the available pool of tree root derived sources of mycorrhizal inoculum.

5. Conclusions

Forests of the western United States have experienced a departure from their historic condition that was spatially evolved and morphologically adapted to experience varying degrees of fire intensity and severity across large geographic landscapes. This condition is true as well in the Blue Mountains of eastern Oregon, where removal of indigenous peoples, removal of late seral trees, as well as introduction of grazing and ingress of human populations into the forests have resulted in over a century of fire suppression (Langston 1995). This novel landscape has prompted managers to reduce the risk of loss due to large-scale wildfire (Covington and Moore 1994), and researchers to propose mechanisms to accomplish this objective (Hessburg et al. 2015).

Among the choices available to federal land managers, the two most commonly found methods for reducing woody fuel loads and accomplishing the objective of forest restoration are mechanical thinning and prescribed burning. While each has its own unique advantages, each also has its individual drawbacks. Despite a long history of conflicting agendas and mistrust, the emergence and evolution of forest collaboratives has provided an increasingly effective mechanism for promoting the interests of stakeholders and creating agreement on restoration activities (Schultz et al. 2012). Among the stakeholders, one concern that is largely agreed on is that forests in their current condition and management are not meeting the expectations of the public. Regardless of the means by which reduction of risk of uncontrolled wildfire, there is an immense amount of work that needs to be done.

The role of fungi in increasing the survivorship of trees following disturbances including fire is becoming more appreciated as a holistic view of forest management evolves. Linking aboveground productivity with belowground processes will be an increasing priority for land managers as forest restoration moves forward. The development of integrated resource management means that collaboration between researchers across disciplines coupled with consideration to the needs of all those affected by the forests management will be a daunting task. Regardless, there is more of a need now than ever before to accelerate the process and be as proactive about preserving intact healthy forests and creating new healthy forests in the future

CHAPTER 2: FUELS TREATMENTS OF PONDEROSA PINE (*PINUS PONDEROSA*) IN THE BLUE MOUNTAINS OF EASTERN OREGON: A MYCORRHIZA PERSPECTIVE

1. Introduction

Fire suppression, among other factors, has contributed to alteration of previously fire-adapted ecosystems to those more at risk of stand replacing wildfire (Covington and Moore 1994). In the last two decades, fire suppression costs have been rising exponentially as the amount of forested land that annually burns has increased (Covington and Vosick 2016). In effect, fire suppression is producing hotter and larger fires by allowing fuel beds to increase disproportionally from historical levels until they inevitably burn. In response to the rising cost of suppression at a receding benefit of that effort, the potential benefits of proactive actions to reduce wildfire severity have received increased attention. Increased emphasis on restoring and maintaining healthy landscapes along with reduction in limitations to fire management (FLAME Act 2009), as well as public education of the ecological benefits of fire, has facilitated heightened interest about the potential for prescribed fire and manual removal of woody materials from forested areas to decrease fire severity.

Historically, forests dominated by ponderosa pine (*Pinus ponderosa* Lawson & C. Lawson) extended throughout much of the western United States (Oliver and Ryker 1990; Graham and Jain 2005). These forests were characterized by frequent fires that removed understory vegetation and promoted cohorts of trees clumped in densities as low as 20-57 trees ha^{-1} (Covington and Moore 1994). This sparse distribution allowed increased levels of light to reach the forest floor, and when combined with frequent fire, promoted an understory primarily composed of grasses and herbaceous plants. With the advent of fire suppression, the janitorial services provided by fire were removed and cascading changes in plant community composition resulted in a shift from herbaceous plant to shrub dominance. This was combined with the encroachment of shade-tolerant and fire-susceptible tree species such as grand fir (*Abies grandis* (Dougl. ex. D. Don) Lindl.) and white fir (*Abies concolor* (Gord. & Glend.) Lindl. ex Hildebr) (Moore et al. 1999). In-growth of this community changed fire interactions within these landscapes, bolstered competitive stress for water and nutrient sources, and left trees more susceptible to attack from pathogens such as bark beetles (Youngblood et al. 2004). Fires that were historically confined to the forest floor became able to climb a vegetative "ladder" that allows fire to enter the crown layer of the forest canopy and move quickly through the forest, resulting in tree mortality outside the historic range (Binkley et al. 2007). To mitigate risks of future stand-replacing wildfires, forest managers have accelerated the use of fuel reduction methods (i.e. prescribed fire and thinning), but do so with little comparative knowledge of their short and long-term environmental effects.

In the Blue Mountains of eastern Oregon, at the original site of the nationwide Fire and Fire Surrogate study (FFS) network (http://www.fs.fed.us/ffs/sites.htm), fuels reduction with low-intensity prescribed fire and mechanical thinning/removal were studied at the operations scale, a novel approach at the time. A limitation of previous research was that many studies were conducted at scales relatively easy to measure, but not applicable beyond a single site (McIver et al. 2000). Research at the first FFS project area, known as Hungry Bob, incorporated research considerations including wildlife, insects, economics, forest pathology, vegetation, fuels, soils, and soil microbial communities (Youngblood et al. 2006; McIver et al. 2013).

The objective of the FFS study was to determine the effects of fuels reduction treatments to transition fires from those of high severity, and often stand-replacing, to those of a low severity. Fire severity is a secondary metric related to fire intensity. Fire intensity relates to the quantitative factors of a fire (temperature, flames height, duration of heat pulse), while fire severity relates more to the qualitative consequences of the fires intensity (amount of vegetation killed, effects on soil strength/properties, damage to roots) (Keeley 2009). High severity fires may burn at soil surface temperatures exceeding 300°C (Smith et al. 2016) and cause partial to total vegetation mortality aboveground and complete or near complete loss of belowground soil microbes in the top 15cm of the soil profile (Rundel 1983; Hebel et al. 2009; Reazin et al. 2016; Smith et al. 2017). In contrast, low severity fires typically experience temperatures below 100°C at the surface and remove mostly smaller shrubs and small diameter trees, leaving larger trees and soil microbes below the top 5cm intact (Agee 1973; Cowan et al. 2016).

Soil microbes tend to congregate in the upper soil profile where nutrient concentrations are highest (Oliver et al. 2015), decrease in frequency with increasing depth (Anderson et al. 2014), and respond quickly to ecosystem disturbances such as fire (Smith et al. 2004, 2005; Barker et al. 2013; Kageyama et al. 2013; Reazin et al. 2016; Smith et al. 2017). However, their long-term response to post-fire disturbance, exacerbated by over a century of fire suppression, is largely unknown. Forests in which low-intensity fire has been excluded have diminished release of nutrients bound in accumulated surface leaf litter, duff, and woody debris (Monleon and Cromack 1996; Monleon et al. 1997). This pool of nutrients, localized in the upper soil layer, creates a chemical gradient that presumably is followed by the belowground microbial community (Hart et al. 2005b). This accumulated duff and organic layer also contributes to increased soil heating in the event of a fire (Ryan, 2002). In a low-severity fire, flames typically have a low duration in any one place, translating into temperatures that barely penetrate the soil, leaving the microbial community intact. Deep organic layers at the soil surface provide fuel that increase the duration of time heat radiates into the soil (Busse et al. 2013; Smith et al. 2016), and have the potential to heat soil to temperatures lethal to soil microbes, which can remain altered after more than a decade (Klopatek et al., 1990).

Soil microbes are directly responsible in many ways for the survival of forest trees. Mycorrhizal fungi exist in a unique symbiotic relationship with most plant families, including the Pinaceae, where they create a sub-category of the symbiosis known as *ectomycorrhizae*. In this relationship, the symbiotic fungal partner expands the functional root network of the tree by growing over an increased surface area, making it possible to access water and nutrients that non-colonized tree roots would be unable to access on their own. In many forested systems, the primary limiting nutrients to tree growth are nitrogen (N) and phosphorus (P). Mineralization (mobilization) of these nutrients is often facilitated by other soil microbes in the interstitial space between soil particles (Binkley et al. 2012) and are then absorbed and transported via the mycelial network to the fungal symbionts host tree. In exchange for nutrients, the host tree provides carbon (C) to the ectomycorrhizal fungi (EMF) in the form of sucrose, which is then converted to glycogen by the fungi (Smith and Read 2010). Additional services of EMF to the host tree include physical and chemical protection from antagonistic/pathogenic fungi (Smith and Read 2010), and connection and transfer of C among other host trees through *common mycorrhizal networks* (i.e. shared mycelial connections among trees) (Molina and Trappe 1982; Molina et al. 1992; Simard et al. 2015).

There is a large pool of research on mycorrhizal fungi response to experimental changes to their environment, but few studies investigate beyond a few growing seasons. Indeed, there is a paucity of long-term research on mycorrhizal responses to fire and fire-mitigation (Bastias et al. 2006; Dooley and Treseder 2012; Holden et al. 2013; Oliver et al. 2015; Overby et al. 2015). It could be expected that after forestry operations to reduce fire risk (mechanical thinning and prescribed fire), adverse effect on fungal populations could occur. Smith et al. (2005) determined this outcome, concluding that following restoration activities, fungal abundance and diversity were significantly decreased two years following thinning and one year following subsequent burning treatments. Some evidence suggests that long-term short rotation burning can alter the belowground EMF to a depth of up to 10cm (Bastias et al. 2006; Hart et al. 2005a). What is less well known is the long-term response of mycorrhizal fungi to restoration practices and if initial damage to the fungal communities creates long-term consequences.

The objective of this study was to assess the EMF community in the dry-climate region of eastern Oregon where fuels reduction treatments were implemented at the Hungry Bob study site over 15 years ago. Specifically, did the impact of mechanical thinning, prescribed burning, or a combined effect of the two compared to untreated forest stands have a transient or long-term effect? Additionally, is there a spatial component to the communities' variation within the top 10cm of soil among each treatment type? Also investigated were differences among treatments in soil nutrients C, N, P, pH and soil bulk density as well as the recovery of litter which contributes to nutrient cycling near the soil surface.

2. Materials and Methods

2.1 Study Area

Samples were collected from the Hungry Bob study area located on the Wallowa Valley Ranger District (Wallowa-Whitman NF) within the 12,000 ha "Waipiti Ecosystem Restoration Project" (Matzka 2003). The Hungry Bob project area is located between the Crow Creek and Davis Creek drainages (45° 38' N, 117° 13' W), about 45km north of Enterprise, OR (Fig. 1), and was designed as an operations-scale experimental group of harvest units approximately 9400 ha (50 km^2) in size for the purpose of evaluating the economic and environmental impacts of restoration timber harvesting and prescribed underburning (Youngblood et al. 2008).

Soils found at the study site are generally derived from ancient Columbia River basalts, which form steep topographies interspersed with numerous plateaus, draws, and ridges. Soil depth varies from deep to shallow depending on aspect and elevation, and soil profiles include Typic Vitrixerands from the Olot series, Vitrandic Argixerolls from the Melhorn and Larabee series, Lithic Ultic Haploxerolls from the Fivebit series, and Lithic Haploxerolls in the Bocker Series (Youngblood et al. 2006). In this area soils generally support plant communities dominated by ponderosa pine and sometimes mixed with Douglas-fir (*Psuedotsuga menziesii* (Mirb.) Franco), grand fir, and some lodgepole pine (*Pinus contorta* Douglas ex Loudon var. *latifolia* Engelm. ex S. Watson). Elevation at the study area ranges from 1040m to 1480m (Youngblood et al. 2008). Annual temperatures average 7.8°C. Annual precipitation averages 50cm, along with an average of 66cm occurring in the form of snow. The two distinct periods of precipitation occur annually, with snow occurring in November to February and rain occurring in March (Youngblood et al. 2006).

Previous to the treatments, the forests surrounding the Hungry Bob were impacted by over a century of fire-suppression as well as timber harvesting and grazing pressure. Timber harvest early in the 20th century resulted in removal of most of the larger trees, and forests in the area are generally in the stem-exclusion phase (O'Hara et al. 1996). Trees found at the study site are predominately second-growth (harvesting occurred as recently as 1986), with post-treatment measurements as follows: tree diameters averaging approximately 33.5cm dbh, and stand basal areas averaging around 26 m^2 ha^{-1}, average SDI of 206, and post-harvest trees ha^{-1} of 26, with some residual large diameter trees that are 100-200 years old (Matzka 2003). The understory consists of the ponderosa pine/snowberry and Douglas-fir/snowberry plant associations (Johnson and Clausnitzer 1992), with vegetation dominated by snowberry (*Symporicarpos albus* (L.) S.F. Blake), white spirea (*Spirea betulifolia* Pall.), pinegrass (*Calamagrostis rubescens* Buckley), Idaho fescue (*Festuca idahoensis* Elmer) and bluebunch wheatgrass (*Pseudoroegneria spicata* (Pursh). The forest floor is covered in a layer of primarily ponderosa pine needles that averages 3cm in depth.

2.2 Experimental Design

From a set of 44 potential stands, 37 were determined to be the most suitable for experimental units in regard to topography and stand structure (Youngblood et. al. 2006). Of these, 16 units were randomly selected for operational experimentation and assigned to treatments for a completely randomized study design. Experimental units varied from 10-20 ha in size. Four replications of each of the four treatment types were randomly assigned to units. The treatments were:

Control- Untreated or no action

Thinning- single entry from below

Prescribed Fire- single underburning of forest floor

Thin + Burn- thinning from below followed by prescribed underburning of forest floor

Thinning was conducted in 1998 via mechanical harvesting systems. Burning was scheduled for 1999 but due to unfavorable weather conditions was delayed until the fall of 2000. Restoration treatments were designed to create stands of trees that would adhere to/exceed the “80/80 Rule”, where if a head fire were to pass through treated stands, 80% of the trees would survive under 80th percentile weather conditions (Weatherspoon & McIver 2000). For thinning treatments, silvicultural prescriptions for the stands were slanted towards establishment of irregular gaps and clumps of fire-resistant ponderosa pine into the dominant and co-dominant size classes. Trees were marked for harvesting by focusing on smaller diameter trees that would leave behind trees that would establish dominant and co-dominant classes. Treatments were designed to reduce total basal area from ~25 to ~16 $m^2 ha^{-1}$ (Youngblood et al. 2006). Felling, limbing, and bucking of trees was accomplished with tracked, single-grip harvesters; yarding was ground based with a wheeled forwarder that used logging residuals left on site to minimize soil compaction.

Burn treatments had individual burn plans developed in order to compensate for site specific characteristics of fuels and terrain (Youngblood et al. 2006). Burning included both backing and strip head fires depending on which would accomplish low flame heights. Ignition was from hand-carried drip torches, beginning in the early afternoons. Thin + Burn treatments followed protocols described for thinning (above) in 1998 and burning (above) in 2000, with the added caveat that burning procedures had to be modified in order to account for full combustion of logging residuals. As stated in Youngblood et al. (2006), units randomly assigned to the no-treatment class were later determined to be dissimilar enough to the treated units (in regard to site characteristics and disturbance history) that they should not be considered “true” controls. Therefore, the use of “control” in referring to the no-treatment units is for simplicity.

2.3 Plot establishment and re-establishment

Individual experimental units were located within stands of trees previously established as part of the operations scale design. Some experimental units comprised the entirety of the stand itself, in most cases it encompassed a portion of a larger stand. In all cases the experimental unit received treatment equally to the rest of the stand. Prior to any treatment, plot networks were established for later sampling by creating grids of points all 50m apart using a staff compass and no less than 50m from stand edge to address edge-related issues. At time of establishment, points were permanently marked with GPS units and staked with either wooden stakes in control and thin-only units (to prevent damage to harvest equipment and reduce costs) or metal posts in burn-only and thin + burn units (to relocate plots immediately following treatment). At the time of re-sampling for the current study, GPS coordinates taken in 1998 in some cases were no longer accurate due to technology advances, or wooden stakes placed previously had decayed or otherwise been destroyed. Therefore, plots had to be re-established within each stand. Using previous GPS coordinates and aerial photo map overlays, plot networks were surveyed until plots from original sampling were found, marked with flagging, and new GPS points recorded (Table 10).

2.4 Soil Sampling

Within each experimental unit, three plots had been randomly selected at the time of the original sampling in June 1998 (Smith et al. 2005) and were re-sampled for the current study. Within each 0.2ha circular plot, a randomly pre-determined ponderosa pine tree over 20cm DBH was used for sampling. In cases where that tree had died or could not be located, the nearest ponderosa pine to plot was used. Each tree was followed from the base due east until the edge of the canopy (dripline) was reached. At dripline, a 20x20cm square was cut through the O-horizon, litter (Oi) was removed but the duff (Oe) was left intact. Litter depth was measured to the nearest 0.5cm (Table 10). Two separate soil cores were taken at each plot for analysis of soil chemistry and mycorrhizas, respectively. A slide-hammer with a 5cm x 15cm coring head lined with three 5cm sleeves was used to extract soil (AMS Inc., American Falls, Idaho). Each core was separated so the 0-5cm and 5-10cm depth horizons were isolated. Soil was placed into Ziploc™ bags and stored on ice until brought to the lab for cold storage at 0°C.

2.5 Soil Chemistry Analysis

Soil chemical analysis was performed by the Central Analytical Laboratory (CAL) at Oregon State University. Prior to analysis, soils were dried at 105°C for 24 hours, sieved to 2mm, homogenized in a coffee grinder, and 40g packaged in coin envelopes. Samples were analyzed for total carbon (C) and total nitrogen (N) using a dry combustion process utilizing an Elementar vario MACRO Cube. Bray-phosphorus (Bray-P) was determined using sodium bicarbonate extraction following protocols by Olsen et al. (1984) and modified by Horneck et al. (1989). pH was determined using a 1:2 soil to water ratio on an ATI orion PerpHecT Log R metere model 350 following Horneck et al. (1989).

2.6 Fine Root Processing of Mycorrhizas

After being removed from the freezer, each sample was first soaked for no more than 12 hours at 4°C to loosen soil particles. Samples were then rinsed with tap water through a 2mm sieve to remove soil. Fine roots were isolated and kept in petri dishes for examination via stereomicroscopy. Roots were examined with a stereomicroscope (Zeiss Stemi SV6, Jena, Germany), at 40x or higher. Fine roots were examined for presence of mycorrhizal colonization and classified by morphotype following criteria as described in the Colour Atlas of Ectomycorrhizae (Agerer 1997). Group morphotypes were kept in individual PCR tubes and stored in 2x cetyltrimethylammonium bromide (CTAB) at 0°C until molecular analysis.

2.7 Molecular Analysis

From each group morphotype, one root tip was selected for DNA extraction and amplification. Each root tip was pulverized with a micro-pestle and then DNA was extracted using a Sigma Extract-n-Amp kit™ (Sigma Aldrich, St. Louis, Missouri). The extracted DNA was then amplified via PCR of the internal transcribed spacer region (ITS) using primers ITS 1F and

ITS 4 (White et al. 1990). Individual PCR reactions contained 17.65µl of molecular grade water, 7.5µl of 5x PCR Buffer, 3µl of 10x deoxynucleotide triphosphates (dNTPs), 0.85µl of MgCL, 0.4µl of bovine serum albumin (BSA), 0.3µl of each primer, and 0.5µl of DNA template. PCR cycling parameters consisted of a 2-minute denaturation at 95ºC followed by 30 cycles of 94ºC for 30 seconds, 50ºC for 1 minute, 72 ºC for 1.5 minutes, and a final extension of 72ºC for 10 minutes. PCR products were visualized under UV light on a 2.5% agarose gel treated with Gel-Red™ (Biotium, Hayward, California). Positive and negative controls ensured target DNA amplification and non-contaminated reagents. When a sample failed to amplify, another root tip from the same morphotype was used for molecular analysis. DNA from PCR products were purified using ExoSAP-IT® (Affymetrix, Santa Clara, California) and quantified for sequencing using a Qubit fluorometer (Invitrogen, Carlsbad, California).

Purified PCR samples were shipped to the University of Kentucky Advanced Genetics Technology Center (UK-AGTC) for sequencing. Direct sequencing of PCR products was performed by Sanger Reaction using the ABI Big Dye terminator v.3.1 cycle sequencing kit (Applied Biosystems, Foster City, California) on an ABI 3730xl DNA Analyzer. ITS 1F primer was used to sequence PCR products in the forward direction. DNA sequences were compiled and analyzed using Geneious® v6.1.7. Regions with primer motifs of more than 4% chance of error per base were trimmed from sequences. Once trimmed, sequences of high similarity (>99%) were placed into identical groupings (contigs). Operational taxonomic units (OTUs) were defined as those sequences having ≥ 98% similarity to each other. Sequences were analyzed for taxonomic placement using the blastn suite of the Basic Local Alignment Search Tool (BLAST) from the National Center for Biotechnology Information (NCBI) database (Altschul et al. 1997). The morphotyped root tips were given names to the lowest taxonomic level possible based on

BLAST results and the following criteria: OTUs with ≥ 98% similarity over at least 500 base pairs were assigned to the species or species group level; at the genus level, OTUs generally had 96% to 97% similarity, similarities in the 90% to 95% range tended to group by family, and OTUs with similarities in the 80% to 94% range were assigned to an order if the top 5 matches were consistent at that level.

2.8 Statistical Analysis

Univariate soil nutrient data, species count data, and species abundance data were analyzed in RStudio v.0.99.491. Analysis of statistical assumptions yielded data that were all normally distributed except for total C and total N which were naturally log-transformed for statistical analysis and then back-transformed for ratio inference of treatment comparisons. An extension of a two-way analysis of variance was used that included a repeated measures (autoregressive 1) linear mixed model except for litter data that did not have a repeated measures element. Variation from sample units and depth were accounted for in the models. Mean soil nutrient differences and median ratios of differences for log-transformed data and the subsequent confidence intervals were estimated from the model.

Non-parametric multivariate comparisons of species abundance and soil nutrient data were conducted using PC-Ord™ software version 6.2 (McCune and Mefford 2009. Non-metric multidimensional scaling (NMS) ordinations were used to provide graphical representations of variation in community structure among treatments while accounting for soil nutrient data. Species with only 1 occurrence were removed to increase the chance of detecting trends. The analysis used Sørenson (Bray-Curtis) distance measures, run in auto-pilot, penalizing for ties, in "slow and thorough" mode, used 250 runs of real data and 249 runs of randomized data, to produce an NMS output with a stable 2-axis solution that had real and random stress values of 14.52 and 19.8, respectively, and passed the Monte Carlo significance test (p=0.012). Multi-response Permutation Procedure (MRPP) was used to test if there was a difference among communities based on treatment type. MRPP analysis provides a p-value for a test of the hypothesis of no difference between groups and an A statistic that represents the effect size of random chance, with A=0 meaning that significant results are no more or less due to chance, and A=1 means that all sample units within each group are identical.

3. Results

Confidence intervals are presented only for significantly different results. For a complete list of significance values and confidence intervals, refer to the Table associated with the appropriate response variable.

3.1 Soil Physical Properties

Litter

There was no evidence that litter depth differed among treatments ($F_3 = 0.36$, $p = 0.78$) (Tables 1 & 6, Fig. 7).

Bulk Density

For bulk density, there was no evidence of a treatment level effect ($F_{3,12} = 1.93$, p =0.17), but significant evidence of an effect of depth ($F_{1,12} = 24.48$, $p < 0.01$) (Tables 1 & 7, Fig. 8). There was no evidence of an interaction between treatment type and depth ($F_{3,12} = 0.75$, $p = 0.54$). Mean upper layer soil bulk density was estimated to be 0.06 g/cm^3 less than lower soil cores (p = 0.03, DF = 12, CI = -0.08 to -0.02).

3.2 Soil Chemical Properties

Carbon

There were significant differences in C among the treatments ($F_{3,12} = 4.02$, $p = 0.03$) and between depths ($F_{1,12} = 136.02$, $p > 0.01$) (Tables 1 & 2, Fig. 3) but no evidence of an interaction between treatment type and depth ($F_{3,12} = 2.71$, $p = 0.09$). There were no statistical differences in C among treatments compared to the control (Table 2). Among treatments, median soil C content was estimated to be 1.75 times higher in the Thinning treatment compared to the Burn treatment ($p = 0.02$, DF = 12, CI = 1.21 to 2.56) and 2.07 times higher in the Thinning treatment compared to the Dual treatment ($p \leq 0.01$, DF = 12, CI = 1.41 to 3.02).

Nitrogen

Although there was no statistical difference among treatments of median soil N content in the main ANOVA ($F_{3,12} = 2.61$, $p = 0.1$), there was strong evidence that average N differed between soil depths ($F_{1,12} = 98.67$, $p < 0.01$) (Table 1, Fig. 4). There was no evidence of an interaction between treatment type and depth ($F_{3,12} = 0.91$, $p = 0.46$). Median N was estimated to be 1.99 times greater in the upper soil layer than the lower ($p = 0.009$, DF = 12, CI = 1.55 to 2.55). Though not suitable for inclusion of significant trends due to a non-significant main ANOVA value, pairwise comparisons indicate that median N is estimated to be 1.62 times higher in the Thinning treatment compared to the Dual treatment ($p = 0.02$, DF = 12, CI = 1.18 to 2.24) (Table 3).

*Phosphorus (*Bray*)*

For plant available P there was strong evidence of a treatment level effect ($F_{3,12} = 4.97$, $p = 0.02$) as well as a depth effect ($F_{1,12} = 42.57$, $p < 0.01$) (Tables 1 & 4, Fig. 5) and no evidence of an interaction between treatment type and depth ($F_{3,12} = 1.71$, $p = 0.22$). Mean Bray P was estimated to be 24.38 Mg/kg soil greater in the Thinning treatment compared to the Control treatment ($p = 0.01$, DF = 12, CI = 9.68 to 39.08 Mg/kg soil), 26.38 Mg/kg soil greater in the Thinning treatment compared to the Burn treatment ($p \leq 0.01$, DF = 12, CI = 12.82 to 39.94 Mg/Kg soil), and 17.62 Mg/kg soil greater in the Thinning treatment compared to the Dual treatment ($p = 0.03$, DF = 12, CI = 4.05 to 31.18). Mean Bray P content was estimated to be 14.68 Mg/kg soil greater in the upper soil layer than the lower ($p = 0.06$, DF = 12, CI = 6.66 to 22.72 Mg/kg soil).

pH

For pH there was evidence of a treatment level effect ($F_{1,12} = 5.92$, $p = 0.01$) but no difference between depths ($F_{1,12} = 0.06$, $p = 0.81$) (Tables 1 & 5, Fig. 6), and no evidence of an interaction between treatment type and depth ($F_{3,12} = 1.42$, $p = 0.28$). pH was estimated to be 0.45 units lower in the Thinning treatment compared to the Dual treatment ($p = 0.01$, DF = 12, CI = -0.45 to -0.23).

EMF Soil Nutrient Correlations

The NMS ordination, based on sample units in species space, provided a stable two-axis solution (Fig. 13). The axes accounted for a total of 85.4% of the variation among sample units based on the species abundance, with axis 1 and 2 accounting for 63.1% and 22.2% of variation, respectively. Sample unit points that are closer to each other share a higher proportion of species similarity. Graphically grouping the sample units by treatment in the ordination space demonstrated moderate overlap (Fig. 13). None of the treatment types were completely isolated from the others. A phosphorus gradient was correlated with the Thinning and Dual treatments, aligning well with axis 1. Axis 2 showed alignment with pH, and was correlated more with the Control treatment than other treatments overall.

3.3 EMF Communities

Species Diversity

A total of 174 purified PCR samples were submitted for sequencing. Of these, 60 sequences were successfully assigned Family level designation or lower. When sequences within a core were the same species, they were combined to create “contigs” for subsequent root weight abundance measurements. There were a total of 117 taxa among treatments and depths, with 49 taxa (42%) in the upper soil depth and 68 taxa (58%) in the lower depth. Only 15 taxa occurred in two or more treatment units (Fig. 11), 45 (38%) taxa were detected from only 1 plot, indicating that species were widely scattered across the study site. This pattern of a few dominant and a large number of infrequent taxa was also indicated in Smith et al. (2005).

For species richness, there was no evidence of a treatment level effect ($F_{3,12} = 0.41$, $p = 0.25$) or an effect of depth ($F_{1,12} = 2.53$, $p = 0.86$) (Table 1) and no evidence of an interaction between treatment type and depth ($F_{3,12} = 1.13$, $p = 0.61$).

EMF Colonized Root Abundance

For live root biomass, there was no evidence of a treatment level effect ($F_{3,12} = 0.66$, $p = 0.59$) or an effect of depth ($F_{1,12} = 1.06$, $p = 0.32$) (Tables 1 & 8, Fig. 9) and no evidence of an interaction between treatment type and depth ($F_{3,12} = 0.34$, $p = 0.79$).

EMF Community Composition

Multi-Response Permutation Process (MRPP) results yielded no evidence of a difference in EMF composition among treatments (T-stat = -1.66, p = 0.06); test results also had a very low effect size (A = 0.04). None of the individual treatment types differed from each other in pairwise comparison (See Table 9). Indicator Species Analysis showed that *Cenococcum*_1 was correlated with the Control treatment (p-value = 0.02, Indicator Value = 75.0).

4. Discussion

4.1 Project Overview

The results of this study suggest that effects of fuel reducing restoration efforts on the variables measured here are largely short-term, and that recovery of soil biological, chemical, and physical attributes to levels similar to the control occurred within less than 15 years. The only treatment with significant differences from the control was the Thinning treatment, but of particular management interest is that the Thinning treatment in some cases differed from the Burn treatment and consistently differed from the Dual treatment. A 15-year recovery period is well supported as sufficient time for these soil responses to thinning and burning (Binkley et al. 1992; McKee 1982; Hart et al. 2005b; Busse et al. 2009). In a meta-analysis of the Fire Fire-Surrogate study as a nationwide network, Boerner et al. (2009) found that regardless of treatments across multiple landscapes and environments, the impacts to soil physical and chemical attributes were both “modest in magnitude and transient in duration”. At a site similar to the Hungry Bob, also located in the Blue Mountains of eastern Oregon, Hatten et al. (2008)

found no differences in soil C, N, pH or plant-available nutrients 7 years after spring or fall prescribed burning compared to the control.

In discussion of the results, it is important to note that the sampling methods for this study were intentionally biased towards capturing the mycorrhizal community response. Our finding of similar fine root biomass in the upper and lower 5cm cores did not support the hypothesis that greater mycorrhizal abundance would be found in the upper core. However, microbial abundance and colonization is often highest near the soil surface (Oliver et al. 2015) and decreases with depth (Anderson et al., 2014), supporting the finding of Knicker (2007) that mycorrhizas tend to occur mainly in the top 2.5cm. Our methods included exclusion of the loose litter layer (Oi) but *included* the early and late litter de-compositional layers (Oe and Oa respectively). The addition of organic matter to the upper soil cores resulting from inclusion of this litter material will help explain why values for C, N, and P are higher and bulk density is lower in the upper soil cores than would normally be expected.

4.2 Soil Chemical Properties

In analysis of the long-term effects of fuels treatments within the study site, it was determined that while none of the treatments differed in soil biogeochemical and physical responses from the control (with the exception of greater plant available P in the Thinning treatment), treatments occasionally differed significantly in these responses from each other. The effects of the Thinning treatment created soil nutrient conditions that differed from those with a burning component, and differences with the Dual treatment in particular, were pronounced. This finding is likely attributable to the effects of the post-treatment woody harvesting residual debris and how it was managed. In the Thinning treatment, harvesting residuals were left on site to decompose. In the Dual treatment, harvesting residuals were incorporated into the subsequent burning. This management of the harvesting residuals may have resulted in elevated microbial activity from decomposition in the Thinning treatment and due to increased intensity of combustion of residuals, reduced microbial activity due to less organic matter in the Dual treatment. Moreover, annual temperature and precipitation regimes of the eastern Blue Mountains permit for mobilization/leaching of nutrients for a relatively short period each year, which could in part explain why trends created during initial treatment may still be detectable in this study over a decade post-treatment.

Total soil C was found to be highest in the thinning treatment, which aligns with findings by Chatterjee et al. (2008) and Yanai et al. (2003) who hypothesized that long-term increased soil C following harvesting based restoration treatment was likely due to incorporation of forest residuals into the soil layer. Other mechanisms proposed include increased leaf/root turnover from the forb/shrub community (Campbell et al. 2009) and interactions of soil order and time (Nave et al. 2010).

Nitrogen trends generally following C trends closely is supported in this study. Nitrogen mobilization can increase as more labile sources of N are released by decomposition of slash in concert with labile C availability that promotes microbial activity (Grady and Hart 2006; Monleon and Cromack 1996). Johnson and Curtis (2001) report increases of up to 18% of soil C and N could be expected when tree harvesting residuals are left on site. Our detection of increases in N of up to 30% in the Thinning treatment is likely attributable to the incorporation of organic material found within our sampling methodology. Combustion of woody material initially leads to a pulse of N within the first few years of burning (Covington and Sackett 1992; Wan et al. 2001; Schoch and Binkley 1986) but this effect diminishes within a few years (Binkley et al. 2012). Due to lack of further treatment since initial burning and little residual organic matter on the floor of burned units, it is probable that uptake by plants in burned units have left overall N levels lower than the Control and Thinning treatments.

Given that the Control treatment units in this study were atypical compared to other pre-treatment areas in this study, one of the considerations addressed was if there were any existing differences in soil chemical make-up at the time of treatment. An exploratory ANOVA of pre- and post-treatment soil chemistry data revealed that prior to treatment, the Control was not significantly different from the other treatments. However, similar to the current study, measurements indicated a significant difference of soil C in the Thinning treatment compared to the Burn and Dual treatments. This finding indicates there may have been some inherent difference in stand structure or nutrient deposition from the canopy that was driving some of the post-treatment trends seen.

Soil chemical processes are often linked to pH which may in part explain our findings for plant-available P. In this study, plant-available P was the only response variable where the Thinning treatment was significantly different from the Control as well as Burning and Dual treatments. Ash production from combustion of organic material is known to increase soil pH (Neary et al. 1999; Arocena and Opio 2003, Moghaddas and Stevens 2007) due to the higher concentrations of hydroxides in ash and by the process of calcium, magnesium, and potassium displacing hydrogen and aluminum ions in soil. In areas such as the Blue Mountains where precipitation is low, this effect would last for several years. Values for pH were significantly different between the Dual and Thinning treatments (0.45 units lower in the Thinning). In a review of the Fire Fire-Surrogate Network by Stevens et al. (2012), it was determined that even at the network scale, higher pH also occurred in the Dual treatment. Deposition of harvest residuals that were then burned provided the organic material needed to raise pH in the Dual treatment. Conversely, unburned harvest residuals left in the Thinning treatment (Fig. 6) likely contributed to acidification of the soil (Binkley et al. 2012).

Bray-P can be elevated in response to fire. Temperatures exceeding 400°C can mobilize P into orthophosphates that are available for plants utilization (Binkley et al. 2012). However, such extreme temperatures generally are not found in prescribed fire; rarely exceeding 120°C (Cowan et al. 2016), unless induced by practitioners (Smith et al. 2016). In a 20 year interval study of controlled burns in SW Arizona, Wright and Hart (1997) found no differences in mineral P compared to controls. This is consistent with other authors (Binkley et al. 1992; Kaye et al. 2005; Saa et al. 1993) and the study presented here.

4.3 Soil Physical Properties

Soil bulk density did not significantly differ among treatments, even though it is known that prescribed fire can increase soil bulk density (Certini 2005). Bulk density could also be affected by treatment placement on landforms within the study area. Although the treatments were assigned to sample units by complete randomization, the outcome by chance was that units with a burning component were often along ridges where soils were shallow and rocky; Thinning units typically had a lower slope position and deeper soils. Additionally, harvesting equipment in Thinning units operated on trails covered with harvested tree limbs and tops, thus adding low-density organic material to the soil. Litter accumulation (Oi) and subsequent "duff" depth (Oe and Oa) also did not differ among treatments, providing strong evidence that productivity of these sites has recovered well since treatments. Litter recovery is correlated to densities of mycorrhizal root formation (Malajczuk and Hingston 1981) and could in part explain no difference among treatments in mycorrhizal root biomass in this study. Litter depth and fine root biomass have recovered to levels that are similar to those found in the control, contrasting well with findings by Smith et al. (2005), who determined that one year post burning and two years post thinning at this study site, overall mycorrhizal root abundance as well as duff depth was lowest in the Dual treatment.

4.4 EMF Communities

Outcomes from the forest restoration treatments studied here indicate that the ectomycorrhizal symbionts of ponderosa pine (a dominant forest type in northeastern Oregon) responded positively during a period of recovery without further disturbance. Numerous studies point to EMF reductions after disturbance in the short-term (Smith et al. 2004; Smith et al. 2005; Barker et al. 2013) as well as the long-term (Klopatek et al. 1990). The historic fire return interval in the region where this study was conducted is estimated to be every 15-20 years (Heyerdahl et al. 2001). It could be expected that the EMF communities that had developed prior to fire suppression would have been adapted to recovering within this time period. Other studies support this idea, showing community similarity to controls in as little as 6 years after burning in the southeastern United States (Oliver et al. 2015), or recovered communities within 12 years in severely burned boreal systems (Treseder et al. 2004). Covington et al. (1997) estimated 10-20 years before “(metrics studied) stabilize around some long-term mean”. Other authors suggested similar timeframes: two decades (Holden et al. 2013), twelve or more years (Fritze et al. 1993), or at a minimum a decade (Oliver et al. 2015) to fifteen years (Treseder at al. 2004) before soil microbial populations recover to pre-disturbance levels. All of these findings support 15 years as being an adequate period of time for microbial recovery, as was found in this study.

Negative effects from heat disturbance on microbial communities have been shown to be transient and community recovery has been reported (Hart et al. 2005a; Overby et al. 2015). Cowan et al. (2016) suggest that patchiness of burn intensities coupled with rapid hyphal penetration into areas where EMF were extirpated due to fire could be one mechanism for recolonization of areas previously burned. Latent spore banks within the soil have also been shown to facilitate EMF community recovery after fire (Bruns et al. 1995). Though the burning treatments at the Hungry Bob site reached temperatures higher than desired by burn managers (Jane Smith, personal communication 2017), the strip-head type ignition pattern used in the burning treatments produces a mosaic of patchiness, leaving areas of low to high intensity burning as well as unburned areas (Penman et al. 2007). Low-intensity burns can produce lethal temperatures of 60˚C (Dunn et al. 1985) to a depth of a few centimeters in the soil profile (Cowan et al. 2016; Smith et al. 2016). That scenario was likely the case in this study, yet EMF abundance recovered in the Dual treatment, indicating that even with the hottest temperature penetration and duration as postulated by Smith et al. (2005), recovery of EMF can be dramatic.

The vertical partitioning of mycorrhizal fungi was one of the questions of interest in this study, and evidence for EMF to preferentially distribute within the soil profile has been demonstrated by several authors (Beiler et al. 2012; Dickie et al. 2002; Rosling et al. 2003; Scattolin et al. 2008; Tedersoo et al. 2003). Our results, although not statistically significant, showed that raw mean species richness and mycorrhizal root abundance was higher in the lower core than the upper core, especially in the Dual treatment. Statistically the upper partition of the cores was not greater in EMF, so this finding (perhaps warranting further investigation with more intensive sampling) could reflect the more sheltered moisture conditions of the lower soil profile and represent the preferred distribution of colonized EMF tips in a fire-adapted system. Whether or not this could be attributed to litter recovery and the lack of nutrient cycling in this water-limited system that limits decomposition has yet to be investigated. For most soil chemistry and EMF community variables, the largest differences in responses were between the Thinning and Dual treatments. However, soil chemistry response values were highest in the Thinning treatment, whereas ectomycorrhizal fungus abundance and richness were greatest (though not significantly different) in the Dual treatment where soil chemistry values were typically lowest. Although ash deposition has been thought to facilitate recovery of microorganisms in relation to increased pH (Knicker 2007), it could also be that nutrient depleted soils are driving the EMF response. Trees in soils that are rich in N and P have been found to support less abundant EMF (Nilsson et al. 2005; Toljander et al. 2006), presumably because less carbohydrates need to be allocated to root growth for absorption. Fertilizer effects on ectomycorrhizal fungi are varied and complex. Nilsson and Wallander (2003) found that EM fungi produced about 50% less mycelium in the soil after N fertilization of 100 $kgha^{-1}yr^{-1}$ (as ammonium sulfate) for ten years. Kårén and Nylund (1997) found a 49% reduction in EM root biomass associate with N fertilization of 100 $kgha^{-1}yr^{-1}$ (as ammonium sulfate) for six years prior to sampling. Other fertilization effects on EM showed that the addition of P ameliorated negative effects of nitrogen addition alone on mycelia growth of EM fungi (Nilsson and Wallander 2003). Chatterjee et al. (2008) found that long-term increases in soil C resulted in decreased microbial levels as well. Soils that are impoverished of nutrients necessary for tree growth may be driving increased belowground fine root production which in turn increases the abundance of symbiotic soil fungi.

Among the 15 most prevalent (those with the highest constancy across samples) taxa forming ectomycorrhizae in this study, over half reproduce by forming mushroom fruiting bodies. The other form truffle-like fruiting bodies, are cryptic in various substrates, or are non-fruiting (see Fig. 11). The latter species are members of the genera *Cenococcum*, *Tomentella, Rhizopogon, Tuber,* or *Wilcoxina*. These fungi, despite not having airborne spores, are well represented in studies of both short- and long-term responses to disturbance (Visser 1995). It is possible that colonization by these typical early seral species can then persist across many seral stages, especially if the ecosystem is subject to regular disturbance such as fire. With only two exceptions, the top 15 most prevalent taxa of the 60 found in this study were also the most abundant (by root weight) (Figures 10 and 11). Dominance of a few of ectomycorrhizal taxa within species-rich assemblages is common in pine forest systems (Gehring et al. 1998; Grogan et al. 2000; Stendell et al. 1999; Smith et al. 2004; 2005). Ponderosa pine appears to associate with similar EMF assemblages across a fairly broad ecological range, as many of the more prominent species reported in Trappe et al. (2012) from southern Oregon are also found in this study.

4.5 Resiliency of Fire-prone Landscapes and Management Implications

One of the strengths of the nationwide Fire and Fire Surrogate study was that the primary goal was to assess the effectiveness of fuels reduction treatments at what is known as "the operational scale" (Youngblood 2006). This means that treatments were implemented on a size scale that reflected typical management considerations and not at the smaller sizes more common in experimental research, often due to costs of implementation. As mentioned previously, even prescribed fires have inherent patchiness associated with their burning patterns. During treatment operations at the Hungry Bob site, fire managers were instructed to ensure that sampling areas were burned in order to capture the effect of treatment (J.E. Smith, personal communication 2017). In the short-term (one year post-burn), both burning treatments significantly reduced EMF species richness, live root biomass, and litter depth (Smith et al. 2005). The general recovery of EMF communities to levels not differing from the control is encouraging with regard to the long-term effects of fuel reduction treatments.

Carbon, N, and P levels were highest in the Thinning treatment, but EMF response was greatest (though not significantly different) in the Dual treatment where nutrient levels were lowest. There is perhaps a relationship to the demand for the services EMF provide in terms of nutrient acquisition. In this respect, if the management objective is to maintain the highest amount of mycorrhizal presence on the landscape, then incorporating a burning component to a restoration management strategy would be preferred to thinning alone.

Unfortunately, even though a relatively low-cost strategy, prescribed burning alone is a resource sink and does not provide revenue, and difficulty implementing harvesting operations has increased in recent decades. However, the recent development of forest collaboratives has been identified as a potentially beneficial component to accelerating the process of forest restoration. Thinning alone does accomplish the objective of lowering stand densities and modifying stand composition to accelerate development of late seral characteristics (Youngblood 2006) but it does not remove downed wood on the forest floor and as our study has shown, does not promote higher mycorrhizal presence compared to the Dual treatment. The Dual treatment, which removes organic material from the forest floor by combustion during prescribed burning also recoups expenses from revenue generated during the thinning process.

Burning operations conducted in the fall diminish surface layer EMF more than burning conducted during the spring (Smith et al. 2004). Since it is widely recognized that the process of restoring fire suppressed stands in the western United States will require multiple entries into stands (Hessburg et al. 2005; McIver et al. 2013; Spies et al. 2006; Youngblood et al. 2006), it is important to minimize negative impacts during preliminary entries to ensure ecosystem recovery.

Management focusing on mitigation of risks associated with high severity fire as it is relevant to individual stand conditions is needed. The use of thinning activities exclusively is useful for reducing forest crown connectivity and lowering stand densities while elevating lower canopy height by selecting for larger trees with more desirable crown structure. However, thinning alone does not resolve the issue of surface fuels and promotes continued fine root development near the soil surface. Prescribed fire removes fuel from the forest floor and can remove smaller diameter trees by heat girdling, but cannot accomplish goals of broader tree removal from overly dense stands without particularly hot fires that may damage or remove desirable trees as well. Combining the two treatments types with a preparatory thin followed by prescribed burning captures the benefits of both. The results of this study support the use of thinning activities followed by prescribed burning as it relates to positively impacted long-term mycorrhizal responses. If managers seek to reduce the severity of future wildfires in the shortest and most economical manner, the use of the Dual treatment may be the optimal option so long as there are no stand specific conditions meriting an alternative approach.

6. Conclusions

The results of this study indicate that EMF populations are able to re-establish in areas where they had previously declined as a result of initial thinning and burning restoration activities in the Blue Mountains of eastern Oregon (Smith et al. 2005). This finding supports a growing body of evidence that EMF communities are resilient to low-severity disturbance (Hart et al. 2005a; Overby et al. 2015; Cowan et al. 2016), and expands it to include the activities of mechanical thinning and prescribed burning. This research is valuable to land managers as it provides evidence that aggressive restoration treatments have little long-term effect on EMF and certain soil parameters and this new information may provide more viable options for future management. There is an increased need for accelerated restoration over large landscapes to reduce the risk of stand-replacing wildfire. The presence and abundance of EMF were highest in the Dual treatment, suggesting that lower soil nutrients found in association with regular fire return ecosystems may increase the importance of maintaining EMF in these forest types. Thinning alone resulted in soil conditions with higher nutrient values, while thinning followed by prescribed burning yielded lower-nutrient soil.

If managers are faced with resistance to prescribed burning, thinning alone can be an effective way of reducing stand basal area densities and promoting a discontinuous canopy. However, in order to return ecological processes to the fire-suppressed forests of the mountain west, inclusion of a burning component to restoration treatments is essential. Ultimately, objectives of land managers in the future will include the need to create stands of trees that are resistant to the effects of drought and higher fire-risk resulting from climate change. Reduction of competitive stress by reduction of stand basal area will be a primary approach of restoration efforts. Multiple entries to remove live trees will be required.

The findings of this study are limited to fire-suppressed low-elevation stands of ponderosa pine in the Blue Mountains. Further research to evaluate the outcomes presented here in other fire-suppressed forest ecosystems and across elevations would greatly widen the scope of knowledge about the effects of fuel treatments on ectomycorrhizae. The ability to increase application of these insights would benefit land managers who are faced with an uncertain future of fire, but with a certain call to action.

7. References

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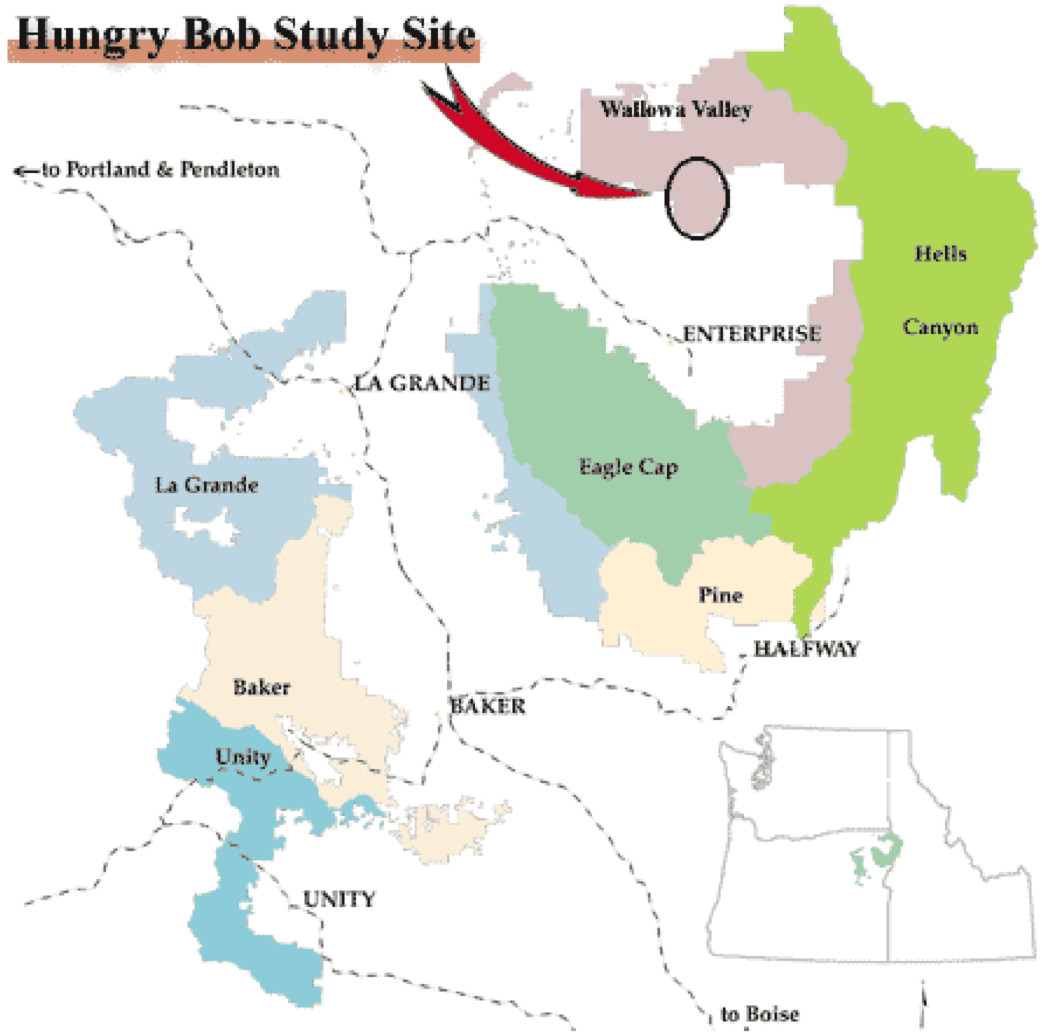

Figure 1. Study Location of the Hungry Bob Experimental Research Area. (https://www.fs.fed.us/ffs/docs/hb/pubs.html)

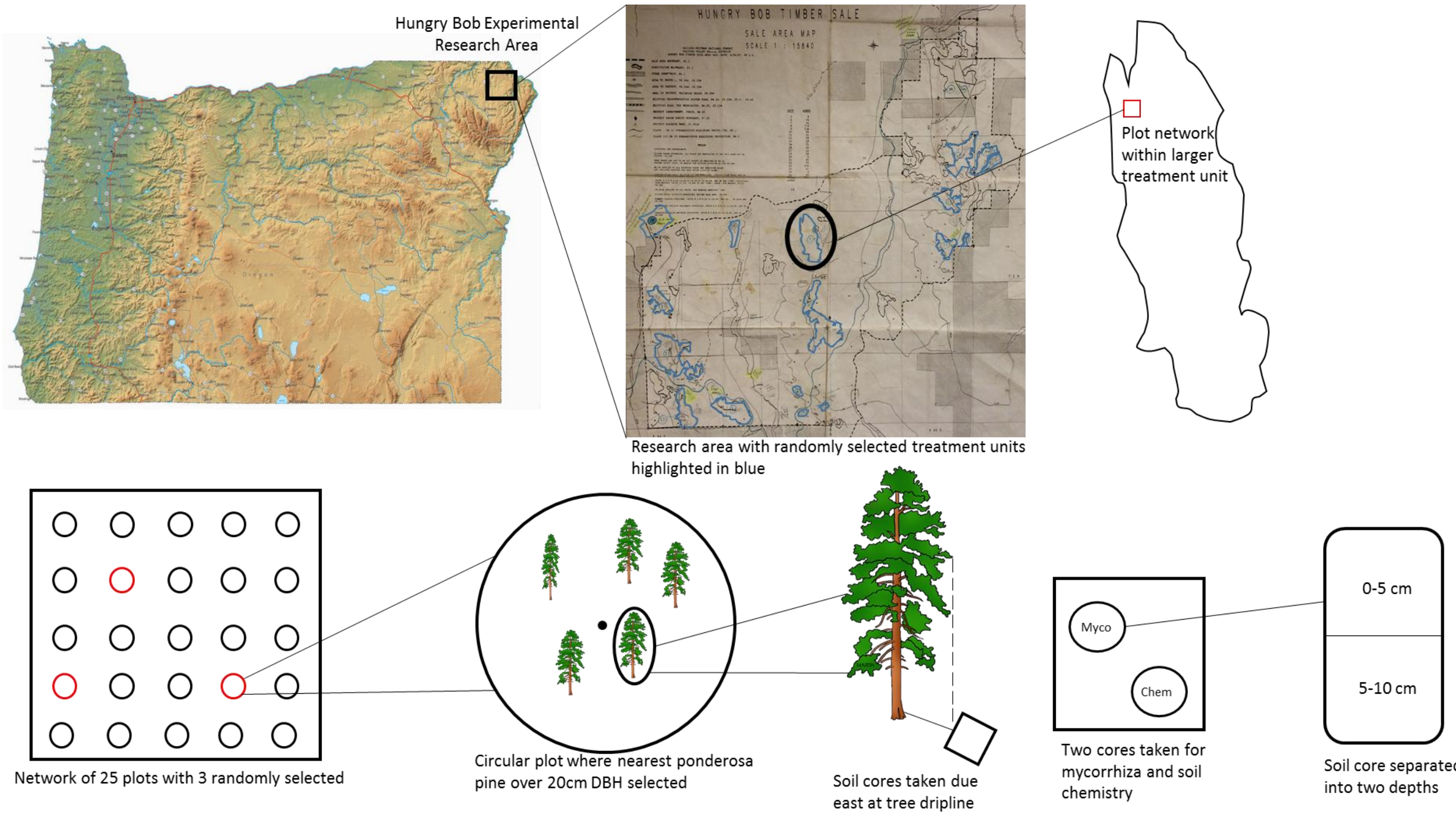

Figure 2. Schematic of sampling location and design.

Table 1. Table of means and standard errors (in parentheses) for mycorrhizal and soil physical/chemical response variables.

Treatment	Depth(cm)	Root Biomass	Species Richness	Carbon (%)	Nitrogen (%)	Phosphorous	pH	C:N	Bulk Density (g/cm^3)	Litter Depth (cm)
Control	0-10	0.06 (0.03)$_a$	3.13 (0.79)$_a$	**5.59 (1.03)**$_{a,b}$	0.29 (0.05)$_a$	**40.14 (2.66)**$_a$	**5.84 (0.05)**$_{a,b}$	**18.95 (0.87)**$_{a,b}$	0.18 (0.06)$_a$	3.31 (1.4)$_a$
Thin	0-10	0.05 (0.01)$_a$	3.75 (0.92)$_a$	**8.96 (2.19)**$_a$	0.39 (0.07)$_a$	**64.52 (3.45)**$_b$	**5.57 (0.07)**$_a$	**21.57 (1.37)**$_a$	0.15 (0.05)$_a$	3.24 (0.43)$_a$
Burn	0-10	0.05 (0.01)$_a$	2.75 (0.92)$_a$	**5.07 (0.91)**$_b$	0.28 (0.04)$_a$	**38.14 (5.34)**$_a$	**5.76 (0.07)**$_{a,b}$	**17.52 (1.11)**$_b$	0.19 (0.05)$_a$	2.92 (0.95)$_a$
Dual	0-10	0.07 (0.01)$_a$	4.34 (0.98)$_a$	**4.24 (0.69)**$_b$	0.24 (0.04)$_a$	**46.90 (6.73)**$_a$	**6.05 (0.05)**$_b$	**17.89 (0.54)**$_b$	0.19 (0.13)$_a$	2.67 (0.93)$_a$
Treatment	**Depth(cm)**	**Root Biomass**	**Species Richness**	**Carbon (%)**	**Nitrogen (%)**	**Phosphorous**	**pH**	**C:N**	**Bulk Density (g/cm^3)**	
Total Upper	0-5	0.05 (0.1)$_a$	4.06 (0.57)$_a$	**8.49 (1.10)**$_a$	**0.40 (0.04)**$_a$	**54.77 (3.79)**$_a$	5.8 (0.08)$_a$	**20.36 (0.84)**$_a$	**0.15 (0.05)**$_a$	
Total Lower	5-10	0.06 (0.01)$_a$	2.93 (0.67)$_a$	**3.44 +/- 0.24**$_b$	**0.19 (0.01)**$_b$	**40.08 (3.81)**$_b$	5.81 (0.05)$_a$	**17.61 (0.59)**$_b$	**0.21 (0.03)**$_b$	
Control Upper	0-5	0.04 (0.02)	3.0 (0.84)	7.87 (1.10)	0.38 (0.05)	44.83 (2.74)	5.85 (0.05)	20.63 (0.22)	0.16 (0.06)	
Thin Upper	0-5	0.05 (0.01)	5.0 (1.14)	13.73 (2.06)	0.55 (0.07)	69.56 (2.24)	5.50 (0.08)	24.43 (0.91)	0.11 (0.03)	
Burn Upper	0-5	0.05 (0.01)	3.75 (0.37)	6.93 (0.92)	0.36 (0.03)	47.14 (4.91)	5.80 (0.11)	18.91 (1.43)	0.17 (0.03)	
Dual Upper	0-5	0.06 (0.01)	4.5 (1.16)	5.54 (0.79)	0.32 (0.04)	57.56 (7.70)	6.05 (0.05)	17.48 (0.24)	0.18 (0.04)	
Control Lower	5-10	0.07 (0.04)	3.25 (1.02)	3.41 (0.18)	0.20 (0.01)	35.45 (1.87)	5.83 (0.06)	17.27 (0.97)	0.21 (0.03)	
Thin Lower	5-10	0.06 (0.01)	2.5 (0.67)	4.19 (0.41)	0.22 (0.01)	59.48 (4.26)	5.64 (0.06)	18.71 (1.08)	0.19 (0.01)	
Burn Lower	5-10	0.04 (0.02)	1.75 (1.35)	3.21 (0.29)	0.19 (0.01)	29.13 (4.84)	5.73 (0.04)	16.15 (0.79)	0.22 (0.05)	
Dual Lower	5-10	0.08 (0.00)	4.25 (1.16)	2.94 (0.25)	0.16 (0.02)	36.25 (4.71)	6.04 (0.06)	18.29 (0.83)	0.22 (0.02)	
BOLD =Indicates at least one treatment significantly differed from the others										

Table 2. Comparisons of soil carbon among treatments and depths.

Response Variable	Overall F-Test Comparison							
Median % Soil Carbon	**ANOVA**	**DF**	**F-Value**	**p-value**	**Statistically Significant**			
	Treatment Type	3, 12	4.02	0.03	*			
	Depth	1, 12	136.02	> 0.01	*			
	Treatment Type : Depth	3, 12	2.71	0.09				
	Pairwise		**t-value**			**Estimate Value**	**Lower C.I**	**Upper C.I.**
	Control : Thin	12	-2.11	0.06		0.64	0.44	0.93
	Control: Burn	12	0.54	0.59		1.12	0.77	1.64
	Control : Dual	12	1.32	0.21		1.32	0.91	1.93
	Thin : Burn	12	2.64	0.02	*	1.75	1.2	2.56
	Thin : Dual	12	3.41	> 0.01	*	2.07	1.41	3.02
	Burn : Dual	12	0.77	0.45		1.18	0.81	1.72
	Summary		**t-value**					
	Thin	12	0.89	0.38				
	Burn	12	-0.37	0.71				
	Dual	12	-0.74	0.47				
	Upper Depth	12	5.6	> 0.01	*			
	Thin : Depth	12	1.79	0.09				
	Burn : Depth	12	-0.22	0.82				
	Dual : Depth	12	-0.94	0.36				
	Pairwise : Depth		**t-value**					
	Upper Control : Lower Control	12	5.61	0.01		2.21	1.71	2.84
	Upper Control : Upper Thin	12	-2.59	0.02		0.57	0.39	0.84
	Upper Control : Upper Burn	12	0.58	0.57		1.13	0.77	1.65
	Upper Control : Upper Dual	12	1.63	0.12		1.41	0.96	2.06
	Lower Control : Lower Thin	12	-0.89	0.39		0.82	0.56	1.21
	Lower Control : Lower Burn	12	0.37	0.71		1.08	0.74	1.57
	Lower Control : Lower Dual	12	0.74	0.47		1.17	0.81	1.7
	Upper Thin : Lower Thin	12	8.15	> 0.01		3.16	2.45	4.06
	Upper Thin : Upper Burn	12	3.17	> 0.01		1.96	1.34	2.86
	Upper Thin : Upper Dual	12	4.24	> 0.01		2.45	1.67	3.57
	Lower Thin : Lower Burn	12	1.27	0.22		1.31	0.89	1.91
	Lower Thin : Lower Dual	12	1.64	0.13		1.41	0.97	2.06
	Upper Burn : Lower Burn	12	5.29	> 0.01		2.11	1.64	2.71
	Upper Burn : Upper Dual	12	1.04	0.31		1.24	0.85	1.82
	Lower Burn : Lower Dual	12	0.36	0.72		1.08	0.74	1.57
	Upper Dual : Lower Dual	12	4.27	> 0.01		1.82	1.42	2.35

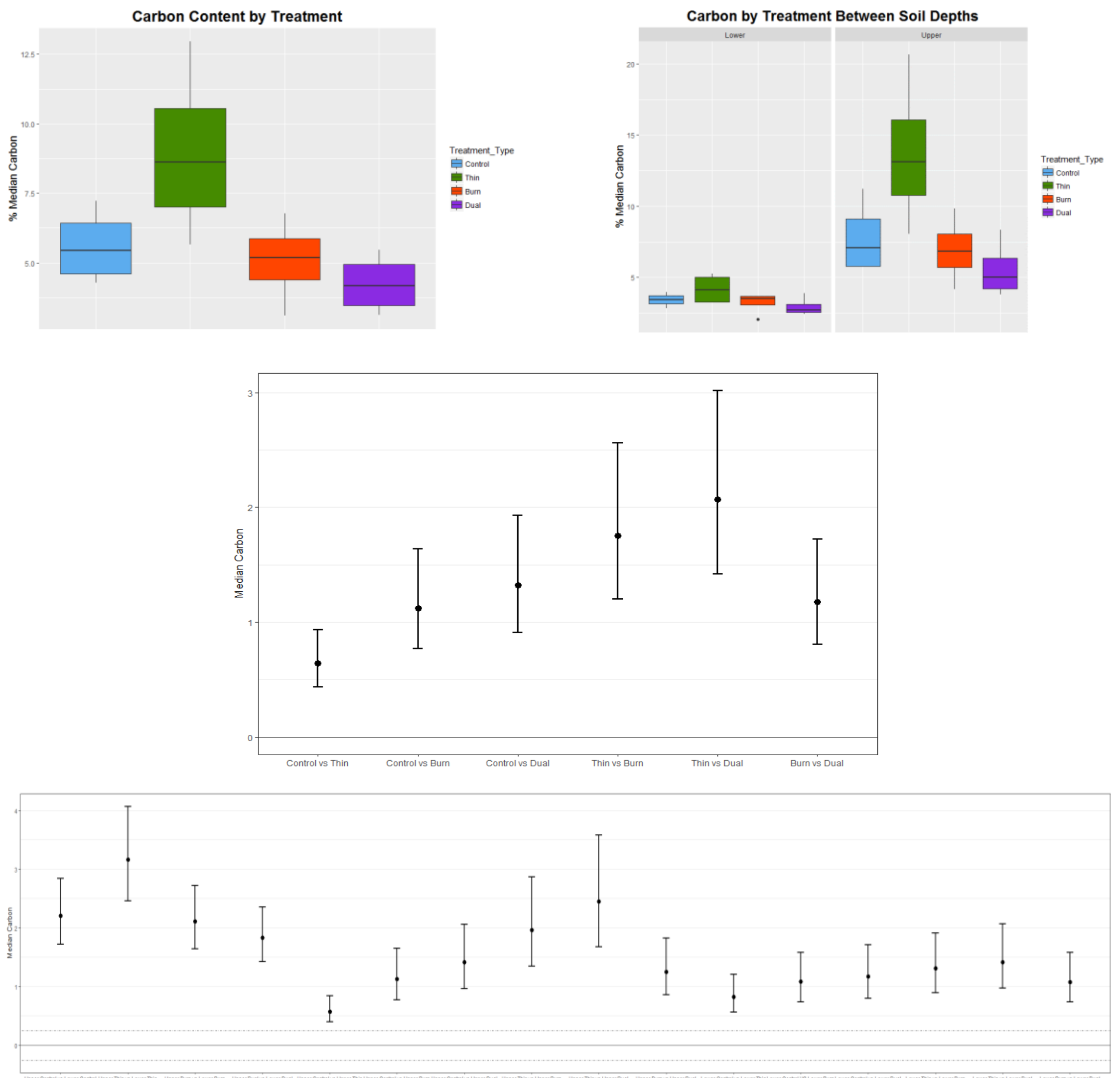

Figure 3. Soil carbon responses at the treatment type level (top left), among treatment types and between depths (top right), estimated confidence intervals of medians at sample unit level (center) and estimated confidence interval among treatment and between depth.

Table 3. Comparisons of soil nitrogen among treatments and depths.

Response Variable	Overall F-Test Comparison							
Median % Soil Nitrogen	**ANOVA**	**DF**	**F-Value**	**p-value**	**Statistically Significant**			
	Treatment Type	3, 12	2.61	0.099				
	Depth	1, 12	95.69	> 0.01	*			
	Treatment Type : Depth	3, 12	0.91	0.46				
	Pairwise		**t-value**			**Estimate Value**	**Lower C.I**	**Upper C.I.**
	Control : Thin	12	-1.71	0.11		0.73	0.53	1.01
	Control: Burn	12	0.11	0.91		1.02	0.74	1.41
	Control : Dual	12	1.01	0.33		1.19	0.86	1.65
	Thin : Burn	12	1.82	0.09		1.38	1.01	1.91
	Thin : Dual	12	2.71	0.02	*	1.62	1.18	2.24
	Burn : Dual	12	0.88	0.39		1.17	0.85	1.61
	Summary		**t-value**					
	Thin	12	0.59	0.56				
	Burn	12	-0.07	0.94				
	Dual	12		0.26				
	Upper Depth	12	4.34	> 0.01	*			
	Thin : Depth	12	1.38	0.19				
	Burn : Depth	12	-0.04	0.96				
	Dual : Depth	12	0.19	0.85				
	Pairwise : Depth		**t-value**					
	Upper Control : Lower Control	12	4.34	> 0.01		1.83	1.43	2.35
	Upper Control : Upper Thin	12	6.31	0.06		0.68	0.48	0.95
	Upper Control : Upper Burn	12	4.28	0.91		1.02	0.73	1.43
	Upper Control : Upper Dual	12	4.62	0.35		1.19	0.85	1.67
	Lower Control : Lower Thin	12	-2.04	0.56		0.89	0.63	1.25
	Lower Control : Lower Burn	12	0.12	0.94		1.01	0.72	1.41
	Lower Control : Lower Dual	12	0.95	0.26		1.24	0.88	1.74
	Upper Thin : Lower Thin	12	2.16	> 0.01		2.41	1.87	3.09
	Upper Thin : Upper Burn	12	2.99	0.05		1.5	1.07	2.1
	Upper Thin : Upper Dual	12	0.83	0.01		1.75	1.25	2.46
	Lower Thin : Lower Burn	12	-0.58	0.51		1.13	0.81	1.58
	Lower Thin : Lower Dual	12	0.07	0.11		1.39	0.99	1.94
	Upper Burn : Lower Burn	12	1.15	> 0.01		1.81	1.41	2.33
	Upper Burn : Upper Dual	12	0.66	0.42		1.16	0.83	1.63
	Lower Burn : Lower Dual	12	1.74	0.3		1.22	0.87	1.71
	Upper Dual : Lower Dual	12	1.08	> 0.01		1.9	1.48	2.44

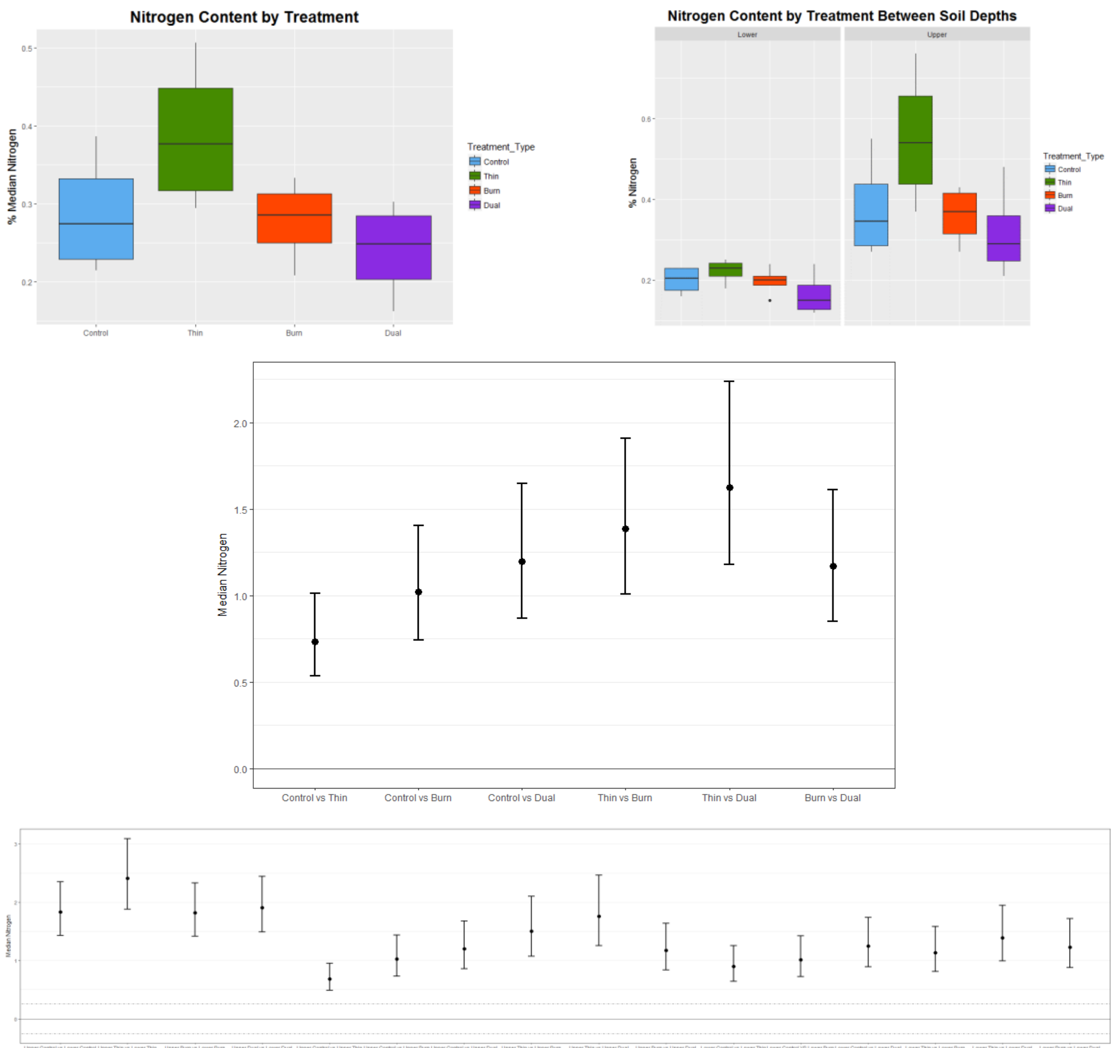

Figure 4. Soil nitrogen responses at the treatment type level (top left), among treatment types and between depths (top right), estimated confidence intervals of medians at sample unit level (center) and estimated confidence interval of means among treatment and between depth (bottom).

Table 4. Comparisons of soil phosphorus among treatments and depths.

Response Variable	Overall F-Test Comparison							
Mean mg P/kg soil	**ANOVA**	**DF**	**F-Value**	**p-value**	**Statistically Significant**			
	Treatment Type	3, 12	4.97	0.02	*			
	Depth	1, 12	42.57	> 0.01	*			
	Treatment Type : Depth	3, 12	1.71	0.22				
	Pairwise		**t-value**			**Estimate Value**	**Lower C.I**	**Upper C.I.**
	Control : Thin	12	-3.21	> 0.01	*	-24.38	-37.94	-10.81
	Control: Burn	12	0.26	0.79		2.01	-11.56	15.56
	Control : Dual	12	-0.88	0.39		-6.76	-20.32	6.79
	Thin : Burn	12	3.46	> 0.01	*	26.38	12.82	39.94
	Thin : Dual	12	2.31	0.03	*	17.62	4.05	31.18
	Burn : Dual	12	-1.15	0.27		-8.77	-22.32	4.79
	Summary		**t-value**					
	Thin	12	2.91	0.01	*			
	Burn	12	-0.76	0.45				
	Dual	12	0.09	0.92				
	Upper Depth	12	2.08	0.06				
	Thin : Depth	12	0.11	0.91				
	Burn : Depth	12	1.35	0.2				
	Dual : Depth	12	1.87	0.08				
	Pairwise : Depth		**t-value**					
	Upper Control : Lower Control	12	2.08	0.06		9.38	1.35	17.41
	Upper Control : Upper Thin	12	-2.99	0.01		-24.72	-39.43	-10.02
	Upper Control : Upper Burn	12	-0.28	0.78		-2.31	-17.01	12.39
	Upper Control : Upper Dual	12	-1.54	0.14		-12.72	-27.43	1.97
	Lower Control : Lower Thin	12	-2.91	0.01		-24.03	-38.73	-9.32
	Lower Control : Lower Burn	12	0.76	0.45		6.31	-8.38	21.02
	Lower Control : Lower Dual	12	-0.09	0.92		-0.81	-15.51	13.91
	Upper Thin : Lower Thin	12	2.23	0.04		10.11	2.05	18.11
	Upper Thin : Upper Burn	12	2.71	0.02		22.41	7.71	37.12
	Upper Thin : Upper Dual	12	1.45	0.17		12.01	-2.71	26.71
	Lower Thin : Lower Burn	12	3.67	0.01		30.35	15.64	45.05
	Lower Thin : Lower Dual	12	2.81	0.01		23.23	8.52	37.93
	Upper Burn : Lower Burn	12	3.99	0.01		18.01	9.98	26.03
	Upper Burn : Upper Dual	12	-1.26	0.23		-10.42	-25.12	4.28
	Lower Burn : Lower Dual	12	-0.86	0.41		-7.11	-21.82	7.58
	Upper Dual : Lower Dual	12	4.73	> 0.01		21.31	13.28	29.33

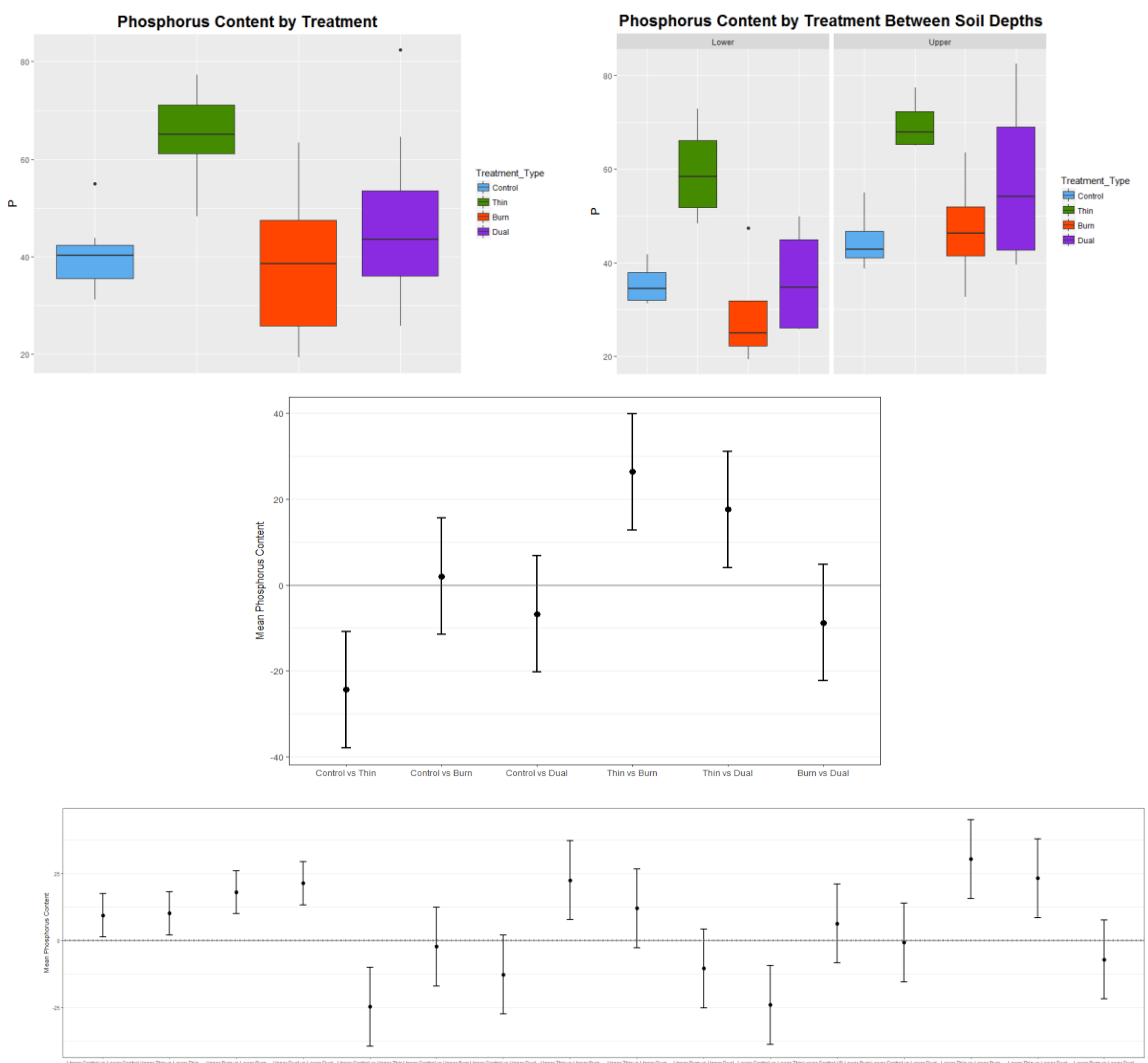

Figure 5. Soil phosphorus responses at the treatment type level (top left), among treatment types and between depths (top right), estimated confidence intervals of means at sample unit level (center) and estimated confidence interval of means among treatment and between depth (bottom).

Table 5. Comparisons of soil pH among treatments and depths.

Response Variable	**Overall F-Test Comparison**							
Mean Soil pH	**ANOVA**	**DF**	**F-Value**	**p-value**	**Statistically Significant**			
	Treatment Type	3, 12	5.92	0.01	*			
	Depth	1, 12	0.06	0.81				
	Treatment Type : Depth	3, 12	1.42	0.28				
	Pairwise		**t-value**			**Estimated Value**	**Lower C.I**	**Upper C.I.**
	Control : Thin	12	1.95	0.07		0.23	0.02	0.45
	Control: Burn	12	0.26	0.79		0.03	-0.18	0.25
	Control : Dual	12	-1.81	0.09		-0.21	-0.43	-0.01
	Thin : Burn	12	-1.68	0.11		0.21	-0.42	0.01
	Thin : Dual	12	-3.75	0.01	*	-0.45	-0.47	-0.23
	Burn : Dual	12	-2.06	0.06		-0.25	-0.46	-0.03
	Summary		**t-value**					
	Thin	12	-1.49	0.16				
	Burn	12	-0.83	0.42				
	Dual	12	1.67	0.12				
	Upper Depth	12	0.26	0.79				
	Thin : Depth	12	-1.47	0.17				
	Burn : Depth	12	0.51	0.62				
	Dual : Depth	12	-0.11	0.91				
	Pairwise : Depth		**t-value**					
	Upper Control : Lower Control	12	0.25	0.79		0.02	-0.12	0.15
	Upper Control : Upper Thin	12	-1.81	0.01		0.35	0.12	0.57
	Upper Control : Upper Burn	12	0.97	0.71		0.05	-0.17	0.27
	Upper Control : Upper Dual	12	0.09	0.14		-0.21	-0.42	0.02
	Lower Control : Lower Thin	12	2.75	0.15		0.19	-0.03	0.41
	Lower Control : Lower Burn	12	0.39	0.42		0.11	-0.12	0.33
	Lower Control : Lower Dual	12	-1.57	0.11		-0.21	-0.43	0.01
	Upper Thin : Lower Thin	12	-2.36	0.09		-0.14	-0.27	-0.01
	Upper Thin : Upper Burn	12	-4.33	0.03		-0.31	-0.52	-0.07
	Upper Thin : Upper Dual	12	-1.97	> 0.01		-0.55	-0.77	-0.32
	Lower Thin : Lower Burn	12	1.49	0.51		-0.08	-0.31	0.14
	Lower Thin : Lower Dual	12	0.82	0.01		-0.41	-0.62	-0.17
	Upper Burn : Lower Burn	12	-1.67	0.34		0.07	-0.06	0.21
	Upper Burn : Upper Dual	12	-0.67	0.07		-0.25	-0.47	-0.02
	Lower Burn : Lower Dual	12	-3.17	0.02		-0.31	-0.54	-0.09
	Upper Dual : Lower Dual	12	-2.51	0.92		0.01	-0.12	0.14

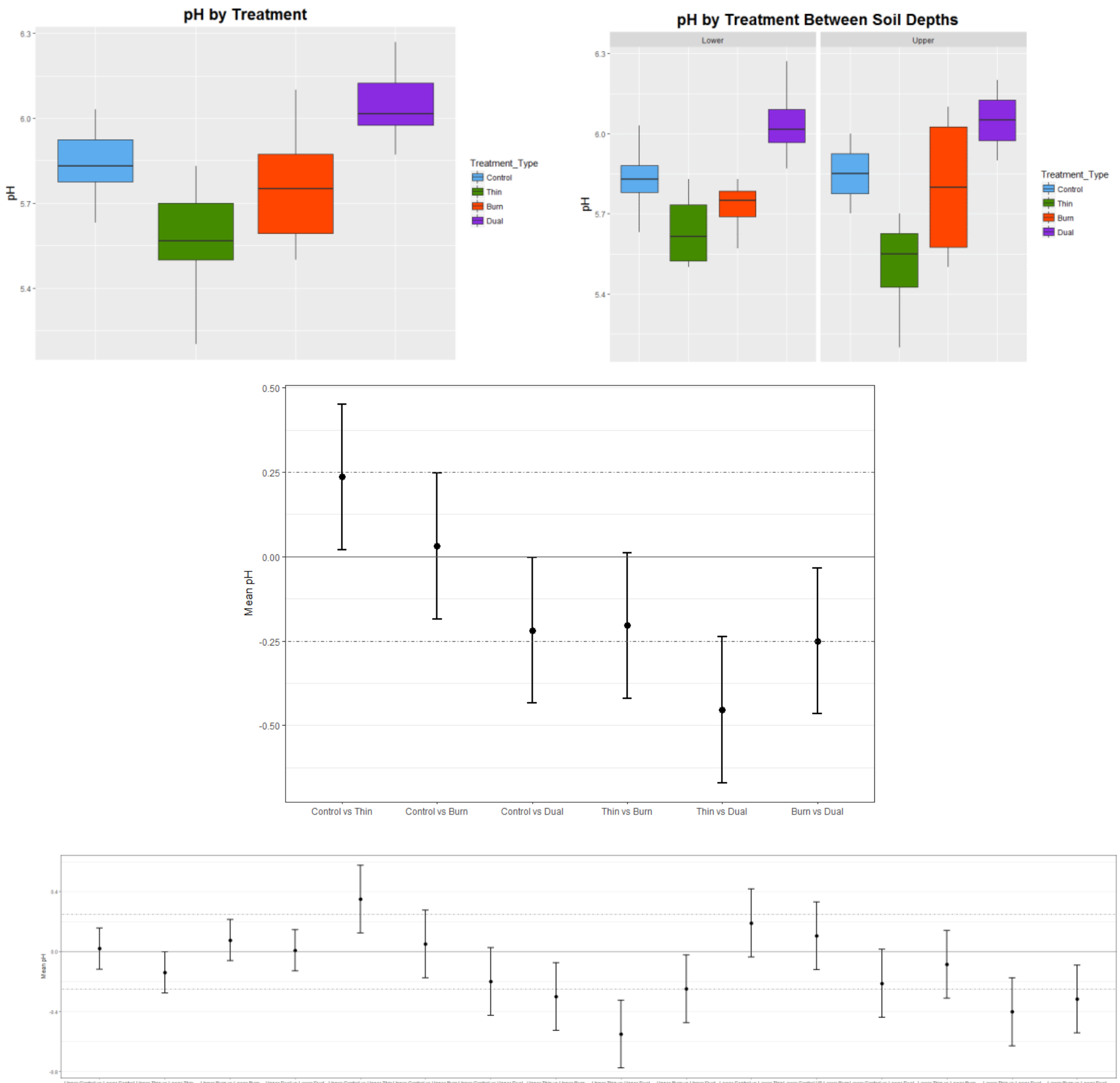

Figure 6. Soil pH responses at the treatment type level (top left), among treatment types and between depths (top right), estimated confidence intervals of means at sample unit level (center) and estimated confidence interval of means among treatment and between depth (bottom).

Table 6. Comparisons of litter depth among treatments

Response Variable	Overall F-Test Comparison							
Litter Depth	**ANOVA**	**DF**	**F-Value**	**p-value**	**Statistically Significant**			
	Treatment Type	3	0.36	0.78				
	All Pairwise		**t-value**			**Estimated Value**	**Lower C.I**	**Upper C.I.**
	Control : Thin	12	0.09	0.92		0.06	-1.17	1.31
	Control : Burn	12	0.54	0.59		0.38	-0.86	1.62
	Control : Dual	12	0.91	0.37		0.64	-0.61	1.88
	Thin : Burn	12	0.45	0.65		0.31	-0.92	1.55
	Thin : Dual	12	0.82	0.42		0.57	-0.66	1.81
	Burn : Dual	12	0.37	0.71		0.26	-0.98	1.51
	Summary		**t-value**					
	Thin	3	-0.09	0.93				
	Burn	3	-0.54	0.59				
	Dual	3	-0.92	0.38				

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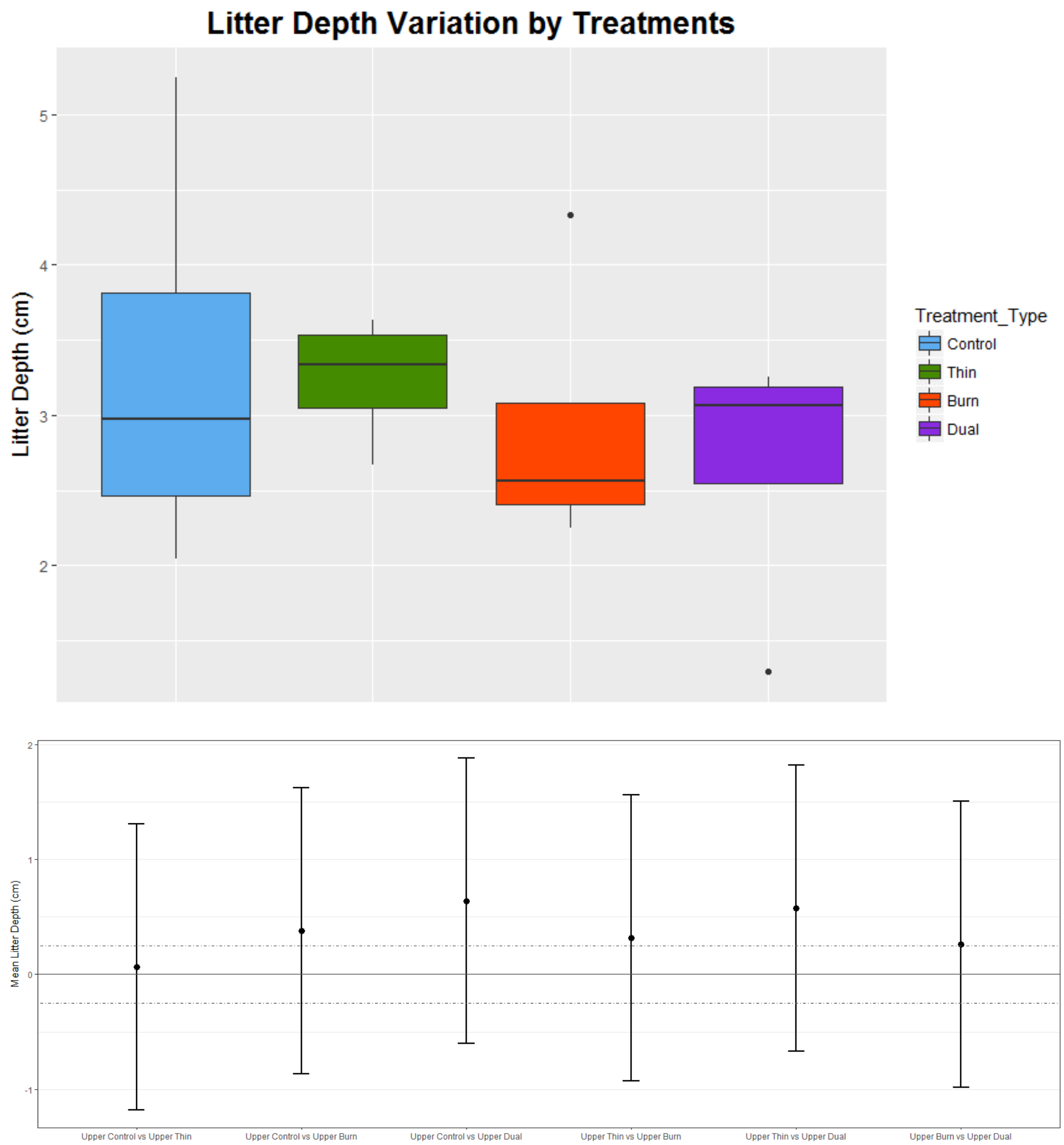

Figure 7. Litter responses at the treatment type level (top left), , estimated confidence intervals of means at sample unit level (center) and estimated confidence interval of means among treatment and between depth (bottom).

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Table 7. Comparisons of bulk density among treatments and depths

Response Variable	**Overall F-Test Comparison**							
Bulk Density (g/m^2)	**ANOVA**	**DF**	**F-Value**	**p-value**	**Statistically Significant**			
	Treatment Type	3	1.94	0.17				
	Depth	1	24.47	> 0.01				
	Treatment Type : Depth	3	0.75	0.54				
	Pairwise		**t-value**			**Estimate Value**	**Lower C.I**	**Upper C.I.**
	Control : Thin	12	1.53	0.15		0.03	-0.01	0.07
	Control: Burn	12	-0.51	0.62		-0.01	-0.05	0.03
	Control : Dual	12	-0.59	0.56		-0.01	-0.05	0.02
	Thin : Burn	12	-2.03	0.06		-0.04	-0.08	-0.01
	Thin : Dual	12	-2.12	0.06		-0.04	-0.08	-0.01
	Burn : Dual	12	-0.09	0.93		-0.01	-0.04	0.04
	Summary		**t-value**					
	Thin	12	-0.68	0.51				
	Burn	12	0.32	0.75				
	Dual	12	0.06	0.95				
	Upper Depth	12	-2.45	0.03				
	Thin : Depth	12	-0.83	0.41				
	Burn : Depth	12	0.14	0.89				
	Dual : Depth	12	0.64	0.53				
	Pairwise : Depth							
	Upper Control : Lower Control	12	-2.45	0.03		-0.06	-0.09	-0.02
	Upper Control : Upper Thin	12	1.71	0.11		0.04	-0.01	0.09
	Upper Control : Upper Burn	12	-0.49	0.63		-0.01	-0.06	0.03
	Upper Control : Upper Dual	12	-0.86	0.41		-0.02	-0.07	0.02
	Lower Control : Lower Thin	12	0.67	0.51		0.02	-0.02	0.07
	Lower Control : Lower Burn	12	-0.32	0.75		-0.01	-0.05	0.04
	Lower Control : Lower Dual	12	-0.04	0.95		-0.01	-0.05	0.05
	Upper Thin : Lower Thin	12	-3.69	0.01		-0.08	-0.13	-0.04
	Upper Thin : Upper Burn	12	-2.21	0.04		-0.06	-0.11	-0.01
	Upper Thin : Upper Dual	12	-2.57	0.02		-0.07	-0.11	-0.02
	Lower Thin : Lower Burn	12	-1.01	0.33		-0.03	-0.07	0.02
	Lower Thin : Lower Dual	12	-0.74	0.47		-0.02	-0.07	0.03
	Upper Burn : Lower Burn	12	-2.25	0.04		-0.05	-0.09	-0.01
	Upper Burn : Upper Dual	12	-0.36	0.71		-0.01	-0.06	0.04
	Lower Burn : Lower Dual	12	0.25	0.81		0.01	-0.04	0.05
	Upper Dual : Lower Dual	12	-1.54	0.15		-0.04	-0.07	0.01

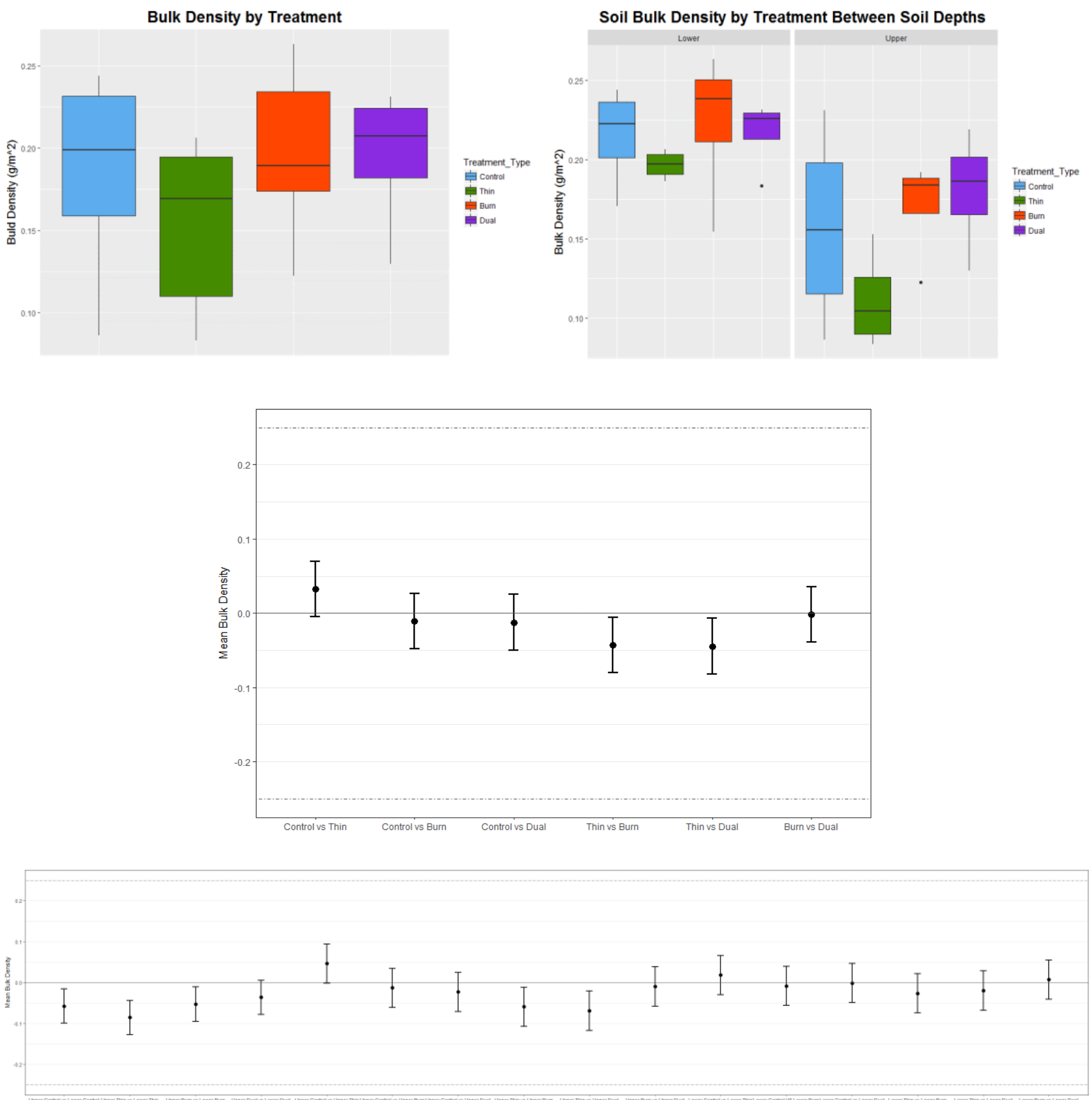

Figure 8. Soil bulk density responses at the treatment type level (top left), among treatment types and between depths (top right), estimated confidence intervals of means at sample unit level (center) and estimated confidence interval of means among treatment and between depth (bottom).

Table 8. Comparisons of root biomass among treatments and depths.

Response Variable	**Overall F-Test Comparison**							
Root Biomass	**ANOVA**	**DF**	**F-Value**	**p-value**	**Statistically Significant**			
	Treatment Type	3, 12	0.66	0.59				
	Depth	1, 12	1.06	0.32				
	Treatment Type : Depth	3, 12	0.34	0.79				
	All Pairwise		**t-value**			**Estimated Value**	**Lower C.I**	**Upper C.I.**
	Control : Thin	12	0.08	0.93		0.01	-0.05	0.06
	Control: Burn	12	0.34	0.73		0.01	-0.04	0.06
	Control : Dual	12	-0.55	0.58		-0.02	-0.07	0.04
	Thin : Burn	12	0.26	0.79		-0.01	-0.04	0.06
	Thin : Dual	12	-0.64	0.53		-0.02	-0.07	0.04
	Burn : Dual	12	-0.91	0.38		-0.03	-0.08	0.03
	Summary		**t-value**					
	Thin	12	0.78	0.45				
	Burn	12	0.66	0.52				
	Dual	12	1.58	0.14				
	Upper Depth	12	0.57	0.58				
	Thin : Depth	12	-0.82	0.42				
	Burn : Depth	12	-0.64	0.53				
	Dual : Depth	12	-0.98	0.35				
	All Pairwise : Depth							
	Upper Control : Lower Control	12	-0.82	0.42		-0.02	-0.07	0.02
	Upper Control : Upper Thin	12	-0.42	0.94		-0.01	-0.06	0.06
	Upper Control : Upper Burn	12	0.64	0.81		-0.01	-0.07	0.05
	Upper Control : Upper Dual	12	-0.58	0.58		-0.02	-0.08	0.04
	Lower Control : Lower Thin	12	-0.07	0.83		0.01	-0.05	0.17
	Lower Control : Lower Burn	12	-0.24	0.41		0.03	-0.03	0.09
	Lower Control : Lower Dual	12	-0.56	0.71		-0.01	-0.08	0.05
	Upper Thin : Lower Thin	12	-0.17	0.67		-0.01	-0.05	0.03
	Upper Thin : Upper Burn	12	-0.49	0.86		-0.01	-0.07	0.05
	Upper Thin : Upper Dual	12	-0.31	0.63		-0.02	-0.08	0.04
	Lower Thin : Lower Burn	12	0.21	0.54		0.02	-0.04	0.08
	Lower Thin : Lower Dual	12	0.83	0.55		-0.02	-0.08	0.04
	Upper Burn : Lower Burn	12	-0.39	0.52		0.02	-0.03	0.06
	Upper Burn : Upper Dual	12	0.61	0.75		-0.01	-0.08	0.05
	Lower Burn : Lower Dual	12	-0.61	0.24		-0.04	-0.11	0.01
	Upper Dual : Lower Dual	12	-1.22	0.56		-0.01	-0.06	0.03

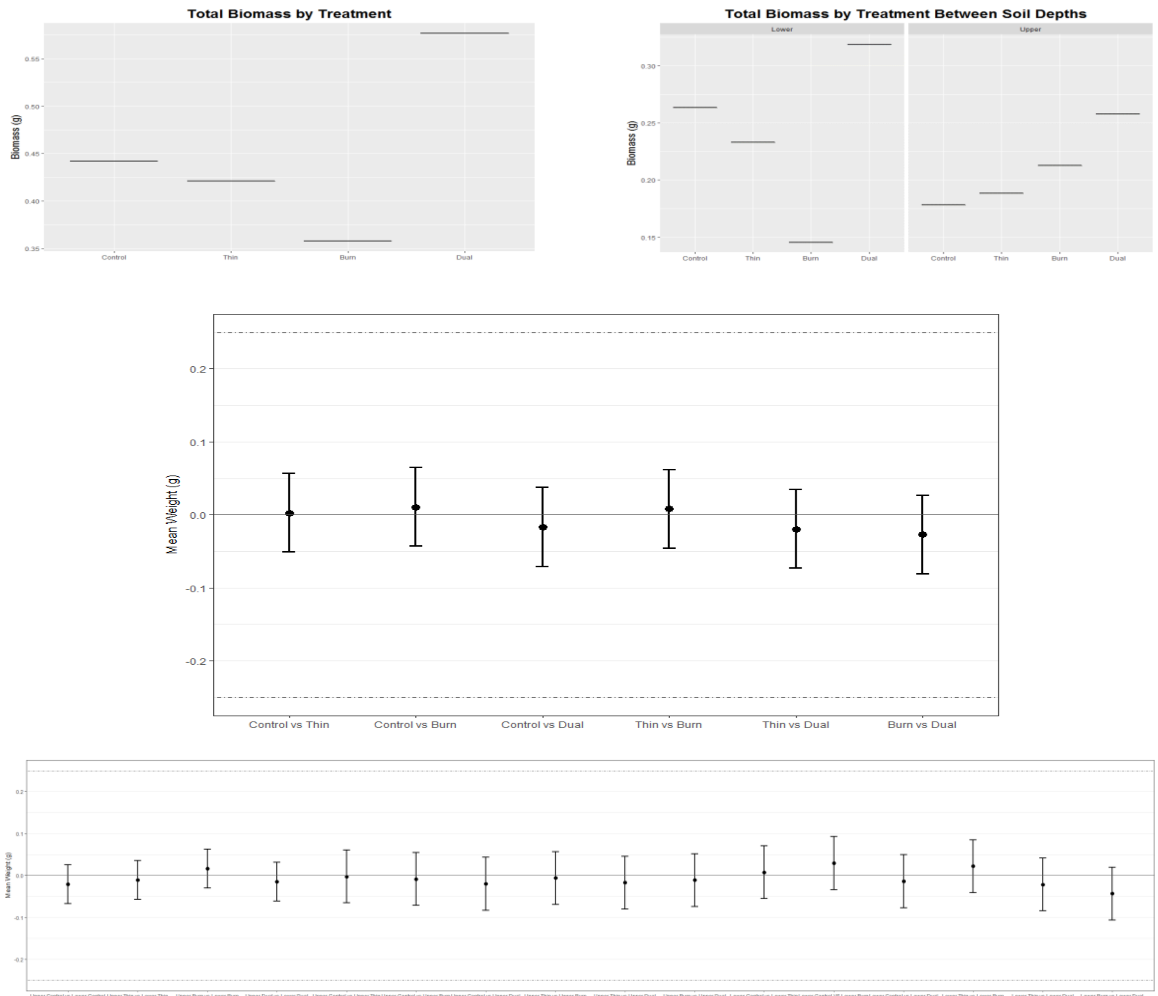

Figure 9. Root biomass responses at the treatment type level (top left), among treatment types and between depths (top right), estimated confidence intervals of means at sample unit level (center) and estimated confidence interval of means among treatment and between depth (bottom).

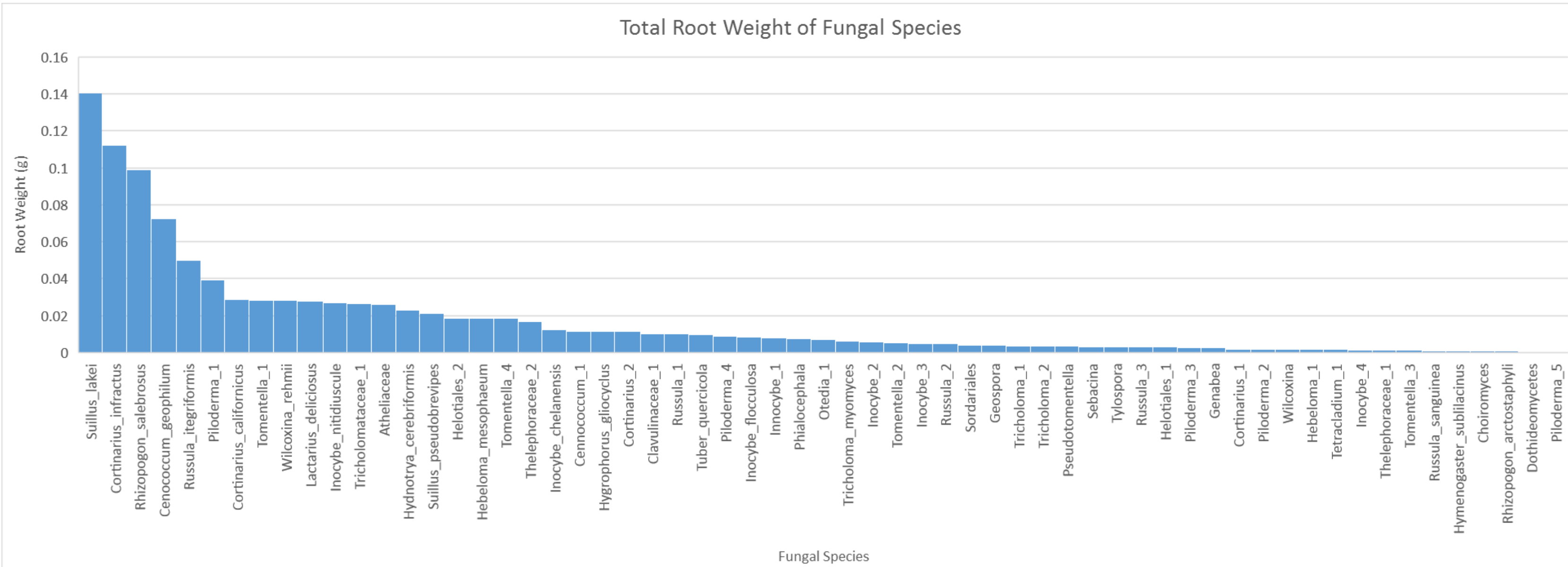

Figure 10. Total Root Weight of Individual Fungal Taxa

Table 9. Comparisons of species richness among treatments and depths.

Response Variable	**Overall F-Test Comparison**							
Species Diversity	**ANOVA**	**DF**	**F-Value**	**p-value**	**Statistically Significant**			
	Treatment Type	3, 12	0.41	0.25				
	Depth	1, 12	2.53	0.86				
	Treatment Type : Depth	3, 12	1.13	0.61				
	Summary		**t-value**			**Estimated Value**	**Lower C.I.**	**Upper C.I.**
	Thin	12	-0.46	0.65		-0.24	-1.33	0.83
	Burn	12	-1.11	0.27		-0.62	-1.81	0.51
	Dual	12	0.53	0.59		0.26	-0.74	1.28
	Upper Depth	12	-0.21	0.84		-0.08	-0.87	0.71
	Thin : Depth	12	1.41	0.16		0.77	-0.31	1.88
	Burn : Depth	12	1.41	0.16		0.84	-0.33	2.07
	Dual : Depth	12	0.26	0.79		0.13	-0.89	1.17

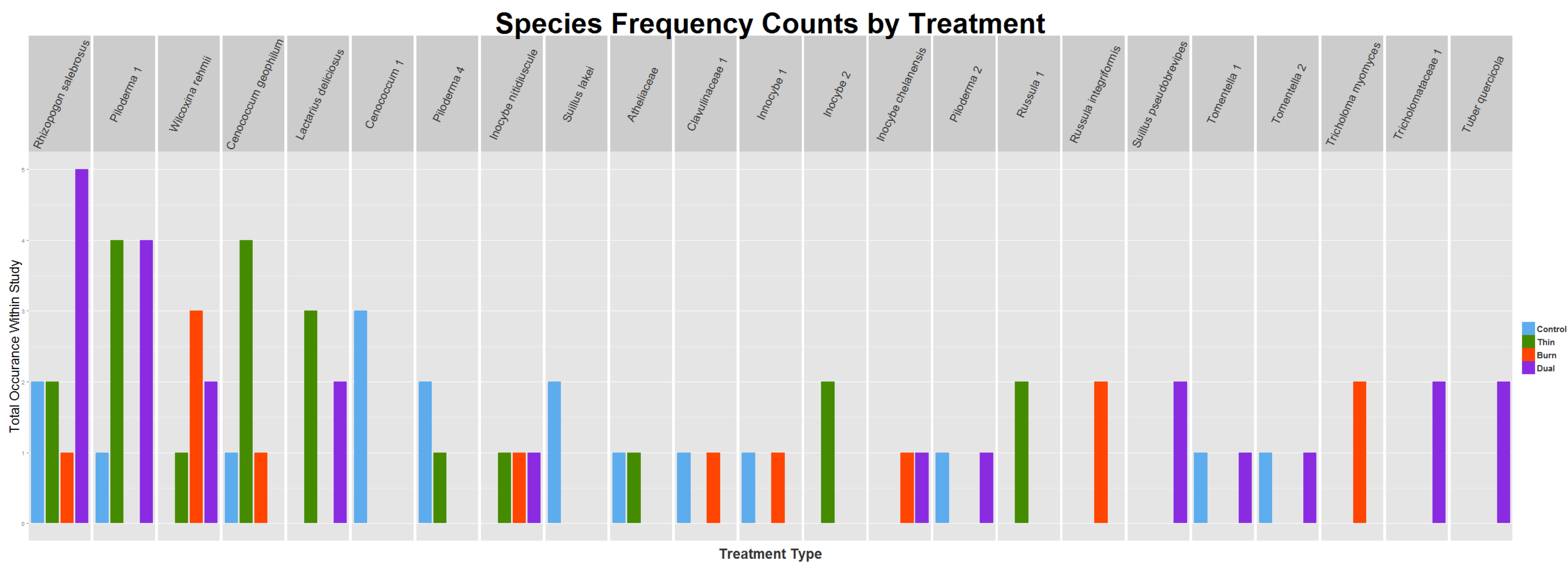

Figure 11. Fifteen most frequent EMF species among all treatment types

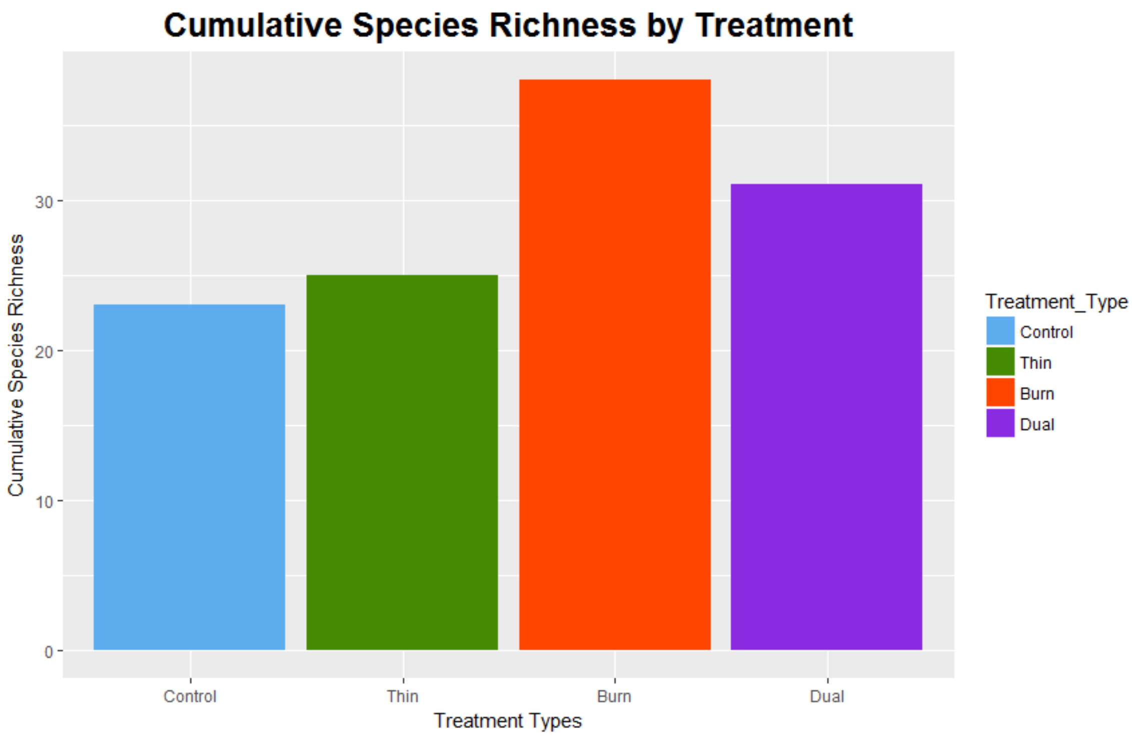

Figure 12. Cumulative species occurrence among treatments regardless of depth. Mean number of species did not differ among treatments (p = 0.25)

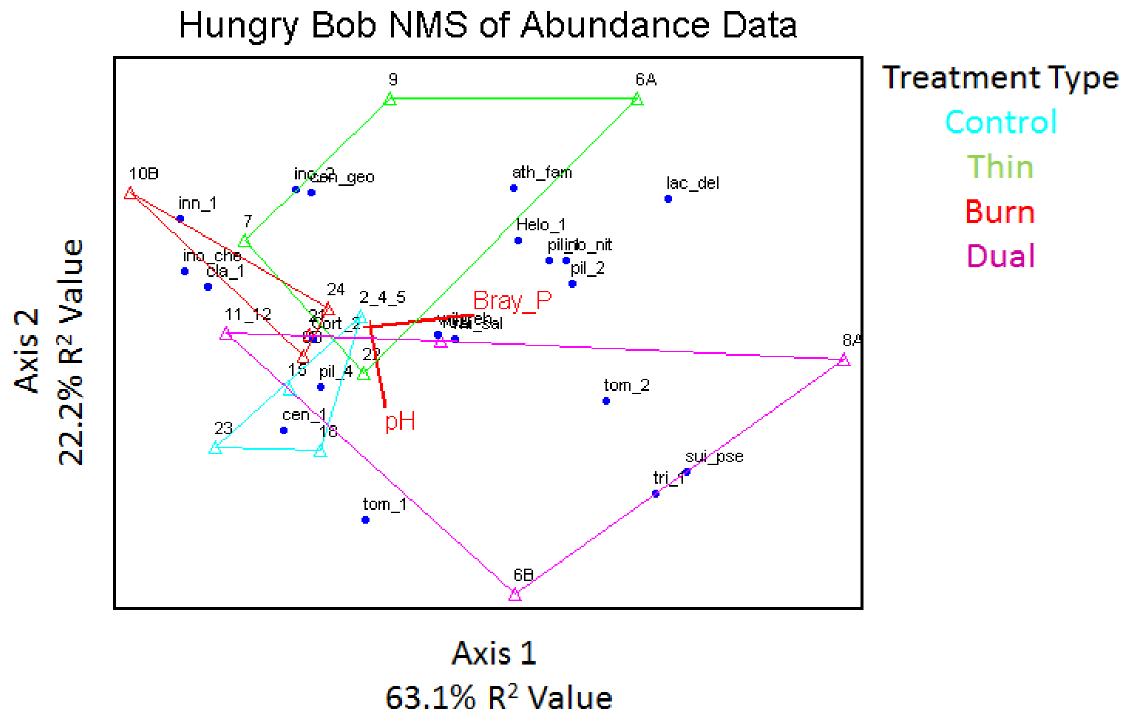

Figure 13. Graphical display of Non-metric Multidimensional Scaling (NMS) ordination. NMS produced according to relative abundance of EMF taxa (n=60 taxa in 16 sample units and 6 environmental variables). Color coded lines represent the region in which sample units are located. R^2 proportion of variance (using the Sørenson distance matrix) among sample units explained by the axes.

Table 10. List of treatment units and the corresponding plots sampled with location UTM.

Unit	Treatment	Plot	lat. (N)	long. (W)	Tree#	2014 Sample Date	2014 Litter (cm)	2014 Litter Avg (cm)
4	Control	4	45.62196	-117.21564	310	20-Jun	3,4,3.5,3	3.38
5	Control	7	45.62552	-117.21777	322	20-Jun	2,2,2,3	2.25
2	Control	6	45.62571	-117.20357	336	19-Jun	2.5,2,2,2	2.22
6A	Thin	9	45.64504	-117.1821368	362	16-Jun	2.5,4,3,2	2.88
6A	Thin	19	45.644517	-117.213417	367	16-Jun	4,2.5,3.5,2.5	3.20
6A	Thin	15	45.642867	-117.2143	372	16-Jun	3,3.5,3.5,4	3.50
6B	Dual	5	45.638667	-117.2109	337	16-Jun	3,4,3,3	3.25
6B	Dual	15	45.6386	-117.210433	353	16-Jun	4,2.5,3,3	3.13
6B	Dual	7	45.63955	-117.2107	345	16-Jun	3,3.5,3.5,2.5	3.13
7	Thin	19	45.658967	-117.21255	3690	16-Jun	2,2,2.5,2.5	2.25
7	Thin	16	45.65971	-117.21421	3684	16-Jun	3,3,3.5,4	3.38
7	Thin	1	45.660133	-117.2123	3673	16-Jun	2,2.5,3,2	2.38
8A	Dual	12	45.68676	-117.17164	590	18-Jun	2,3,3,3	2.75
8A	Dual	21	45.68799	-117.17239	599	18-Jun	3,3,3,4	3.25
8A	Dual	20	45.68752	-117.17308	570	18-Jun	3,4,4,4	3.75
8B	Burn	21	45.68773	-117.16895	566	18-Jun	2,2,2.5,2.5	2.25
8B	Burn	13	45.68753	-117.1695	574	18-Jun	2.5,2,2,3	2.38
8B	Burn	4	45.68669	-117.16748	581	18-Jun	3,3,3.5,4	3.38
9	Thin	6	45.66863	-117.16917	578	17-Jun	7,7,7,5.5	6.63
9	Thin	13	45.66796	-117.16809	581	17-Jun	2.5,1.5,1,2	1.75
9	Thin	18	45.66703	-117.16882	589	17-Jun	2.5,2,2.5,3	2.50
10A	Dual	2	45.67505	-117.15968	700	18-Jun	1.5,2,2,2.5	2.00
10A	Dual	12	45.67405	-117.16162	599	18-Jun	3,3.5,2.5,2.5	2.88
10A	Dual	19	45.67528	-117.16268	377	18-Jun	3,4,5,4	4.00
10B	Burn	11	45.67308	-117.15993	988	18-Jun	3,4,3,4	3.50
10B	Burn	13	45.67231	-117.15993	997	18-Jun	2,1,1,1	1.25
10B	Burn	14	45.67163	-117.15969	788	18-Jun	2,2,2,2	2.00
11	Dual	14	45.65645	-117.17504	746	19-Jun	2,2,1.5,1	1.63
12	Dual	3	45.66371	-117.17141	756	19-Jun	2,2,1,1	1.50
11	Dual	9	45.65603	-117.17375	754	19-Jun	0.5,0.5,1,1	0.75
15	Control	5	45.66268	-117.23525	3647	17-Jun	6.5,7,7,7	6.88
15	Control	3	45.663567	-117.235583	3646	17-Jun	4,5,5,3	4.25
15	Control	1	45.6643	-117.23573	3644	17-Jun	3,5,5.5,5	4.63
18	Control	10	45.66386	-117.25194	737	17-Jun	1,0.5,0.5,2	1.00
18	Control	11	45.66409	-117.25212	766	17-Jun	3,3,3.5,2.5	4.00
18	Control	6	45.662667	-117.254483	777	17-Jun	1.5,1.5,1.5,0	1.13
21	Burn	2	45.64456	-117.25256	783	19-Jun	3,4.5,4,3	3.63
21	Burn	3	45.64493	-117.25288	787	19-Jun	3,2,3,2	2.50
21	Burn	14	45.64735	-117.25476	799	19-Jun	2,2,1,0	1.25
22	Thin	8	45.63401	-117.25343	3602	19-Jun	2,2.5,3,3	2.63
22	Thin	20	45.63412	-117.25382	3610	19-Jun	3,3.5,3,4	3.38
22	Thin	9	45.6342	-117.25299	3616	19-Jun	4,5,4,5	4.50
23	Control	32	45.62097	-117.25075	3620	19-Jun	2.5,2.5,3,3.5	2.88
23	Control	24	45.62019	-117.25126	3625	19-Jun	2.5,3.5,3.5,3.5	3.25
23	Control	14	45.61952	-117.25249	3630	19-Jun	4,3.5,4,4	3.88
24	Burn	1	45.628367	-117.242433	3668	20-Jun	5.5,6,6,5.5	5.75
24	Burn	21	45.62621	-117.24243	3659	20-Jun	3.5,3.5,3.5,3.5	3.50
24	Burn	23	45.62601	-117.24178	3652	20-Jun	3.5,4.5,4,3	3.75

Table 11. List of taxa identified by treatments.

Control	Taxa	Max ID (%)	Accession Number
	Cennococcum_1	96	FJ152538
	Cennococcum geophilum	98	JX630518
	Claulinaceae_1	99	GU180270
	Cortinarius_1	99	JQ393044
	Cortinarius_2	99	FJ039554
	Cortinarius infractus	97	HQ604687
	Geospora	98	EJ188583
	Hebaloma_1	99	DQ822807
	Hebaloma mesophaeum	99	JQ724062
	Hygrophorus gliocyclus	98	EF559269
	Inocybe_1	95	KF617999
	Piloderma_1	95	JQ939098
	Piloderma_2	95	JQ393095
	Piloderma_3	94	JQ393098
	Piloderma_4	99	JQ393093
	Piloderma_5	100	DQ474717
	Rhizopogon arctostaphyli	97	NR121275
	Rhizopogon salebrosus	99	DQ822822
	Sebacina_1	99	JQ393118
	Suillus lakei	98	KJ146731
	Thelephora_1	97	EF655695
	Tomentella_1	96	KM402941
	Tomentella_2	99	JX630568

Thin	Taxa	Max ID (%)	Accession Number
	Atheliaceae	97	EU649086
	Cenococcum geophilum	98	JX630518
	Cortinarius_3	99	JQ393037
	Helotiales_1	90	FJ553817
	Inocybe_2	99	HQ271360
	Inocybe nitidiscule	99	HQ604090
	Lactarius deliciosus	99	EF68508
	Phialocephala	99	JQ711853
	Piloderma_1	95	JQ939098
	Piloderma_2	95	JQ393095
	Piloderma_4	99	JQ393093
	Rhizopogon salebrosus	99	DQ822822
	Russula_1	98	EF218810
	Russula_3	99	KM403067
	Sebacina_2	97	EU819442
	Thelephoracea_2	98	EF619795
	Wilcoxina rehmii	99	AF266708

Burn	Taxa	Max ID (%)	Accession Number
	Atheliaceae	97	EU649086
	Cenococcum geophilum	98	JX630518
	Claulinaceae_1	99	GU180270
	Choiromyces	92	JX630948
	Hydnotrya cerebriformis	99	JF908765
	Inocybe_1	95	KF617999
	Inocybe_3	99	JQ408749
	Inocybe_4	98	HQ604514
	Inocybe chelanensis	99	HQ604403
	Inocybe flocculosa	99	JF908122
	Inocybe nitidiscule	99	HQ604090
	Rhizopogon salebrosus	99	DQ822822
	Russula_2	99	EF218804
	Russula sanguinea	99	EU248591
	Russula integriformis	97	AY310854
	Tetracladium	96	GU327472
	Tricholoma_1	99	KF546500
	Tricholoma myocyces	99	FJ845443
	Wilcoxina rehmii	99	AF266708

Dual	Taxa	Max ID (%)	Accession Number
	Cortinarius californicus	99	FJ039588
	Dothideomycetes	99	EU726287
	Ganebea	98	JQ393052
	Helotiales_2	98	EU649082
	Hymenogaster sublilacinus	98	AF325603
	Inocybe chelanensis	99	HQ604403
	Inocybe nitidiscule	99	HQ604090
	Lactarius deliciosus	99	EF68508
	Piloderma_1	95	JQ939098
	Piloderma_2	95	JQ393095
	Psuedotomentella	90	EU057113
	Rhizopogon salebrosus	99	DQ822822
	Sordariales	99	JN704834
	Suillus psuedobrevipes	97	JN858073
	Thelephoraceae_1	82	EF635793
	Tomentella_1	96	KM402941
	Tomentella_3	91	EF218826
	Tricholoma_2	88	AF377208
	Tuber quercicola	98	JQ393153
	Tomentella_2	99	JX630568
	Tomentella_4	99	JQ393143
	Tricholomataceae_1	98	EU781654
	Tylospora	99	EF619844
	Wilcoxina	98	GU327400
	Wilcoxina rehmii	99	AF266708